Genetic analysis of agro-morphological traits and molecular diversity study (ISSR and SNPs) of quality protein maize (*Zea mays* L.) inbred lines in drought stressed areas of Ethiopia

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(Applied Genetics)

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By

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Addis Ababa University

Declaration

I declare that this thesis is my own original work. It has never been submitted to any other institution anywhere and all sources of materials obtained from other sources have been duly acknowledged in the thesis.

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Lealem Tilahun Amenu Date

Dedication:

To

My uncle

Zerfu Getahun

Source of my inspiration and achievements

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Abstract

Genetic studies along with characterization of elite breeding lines provide understanding of the genetic diversity and relationship among the inbred lines. They also offer information on the type of gene action controlling the inheritance of desirable quantitative traits. The information enables breeders to define a systematic breeding strategy and to select suitable parents and hybrids for further breeding activities and commercialization. Thus, the purposes of this study were to estimate combining abilities and heterosis and to determine genetic variation, correlation, heritability and expected genetic advance of elite quality protein maize (QPM) inbred lines for grain yield and other agronomic traits as well as to investigate their genetic diversity and relationship using SNP and ISSR markers. A total of 116 QPM test cross hybrids developed by crossing 58 QPM inbred lines with two QPM testers was evaluated for 17 morphological traits along with two conventional maize (CM) and two QPM standard checks in drought stressed areas of eastern Ethiopia – Melkassa Agricultural Research Center (MARC), Edo Gojola and Mieso. The inbred lines were also evaluated separately adjacent to the hybrid trial at each site and the inbred lines were genotyped by SNP and ISSR markers. Significant differences were observed among the inbred lines and hybrids for grain yield and most considered agronomic traits indicating that genetic variations existed among the genotypes to allow good progress from selection for improvements of those traits. Across sites, the highest mean grain yield was observed for the hybrid L52/CML159 (5.38 t ha^{-1}) followed by L18/CML159 (5.07 t ha^{-1}) and L35/CML159 (5.02 t ha^{-1}) in the hybrid trial while inbred line L52 showed the highest mean GY which was 3.15 t ha^{-1} followed by L38 (2.94 t ha^{-1}), L47 (2.88 t ha^{-1}), L17 (2.43 t ha^{-1}), L48 (2.33 t ha^{-1}) and L40 (2.06 t ha^{-1}) in the inbred line trial. The combining ability analysis showed that general combining ability (GCA) of lines was significant while specific combining ability (SCA) was non-significant for grain yield, anthesis date, plant height, ear height, plant aspect, ear length and thousand kernel weight indicating that the variability observed among the hybrids was attribuable to additive effects for most traits. The contribution of line

GCA was found to be higher than the contribution of tester GCA and line × tester SCA for all considered traits except for thousand kernel weight where the contribution of tester GCA was higher. Inbred lines L35 (0.83 t ha^{-1}), L45 (0.68 t ha^{-1}), L53 (0.63 t ha^{-1}), L4 (0.57 t ha^{-1}), L21 (0.56 t ha^{-1}), L52 (0.54 t ha^{-1}) and L32 (0.49 t ha^{-1}) had significant positive GCA effects for grain yield. Hybrid combination L52/CML159 had the best SCA effects for grain yield and other most important traits and the maximum standard heterosis over MH140 (19.7%) and MH130 (21.4%). Grain yield had positive and highly significant genotypic and phenotypic correlations with plant height, ear length and thousand kernel weight while negative and highly significant with days to 50% anthesis, anthesis-silking interval, ear position, shoot lodging and ear aspect. The present study showed that root lodging was not important characteristics to be considered while shoot lodging played an important role in determining grain yield. It also showed that ear height is more important than plant height to develop high yielding hybrids and that it is possible to select high yielding varieties which are early but tall with low ear placement. The ISSR markers were found to be as effective as SNP markers in clustering inbred lines into seven sub-groups which are in agreement with pedigree information. All the three multivariate analyses viz. cluster analysis, model-based population structure analysis and principal component analysis using SNPs consistently identified the same seven distinct populations and revealed similar membership of inbred lines in each population. In general, the results from this diversity study based on SNP and ISSR markers will be useful to breeders in selecting best parental combinations for starting new breeding populations, mapping population and marker assisted breeding.

Key words: Quality protein maize, QPM, combining ability, GCA, SCA, heterosis, correlation, heritability, genetic advance, diversity, SNP, ISSR

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List of Abbreviations

AMOVA	Analysis of molecular variance
ANOVA	Analysis of variance
BNMRC	Bako National Maize Research Center
BPH	Better parent heterosis
CIMMYT	International Maize and Wheat Improvement Center
CM	Conventional maize
DAP	Di-ammonium phosphate
EIAR	Ethiopian Institute of Agricultural Research
GA	Genetic advance
GCA	General combining ability
ISSR	Inter simple sequence repeat
MARC	Melkassa Agricultural Research Center
MCMC	Markov Chain Monte Carlo
MoANR	Ministry of Agriculture and Natural Resource
NJ	Neighbor joining
PCA	Principal component analysis
PCoA	Principal coordinate analysis
QPM	Quality protein maize
SCA	Specific combining ability
SH	Standard heterosis
SNPs	Single Nucleotide Polymorphisms
UPGMA	Unweighted Pair Group Method using Arithmetic Averages

1. Introduction

Maize (*Zea mays* L.) has long been, is and continues to be a crop of survival and nutritional security for many resource poor farmers living in most of the developing world (Prasanna, 2013). Its wide adaptability, high yields and valuable by-products are few of the reasons for its popularity. Of 125 developing countries known to produce maize, it is among the three most widely grown crops in 75 of them and it is also preferred as a staple crop by 900 million resource poor consumers (Bekele Shiferaw *et al.*, 2011). In Ethiopia, maize ranks first in terms of total production among all the other cereal crops produced even though second in area coverage next to teff (Tsedeke Abate *et al.*, 2015). According to CSA (2017), out of 16.3 million private peasant holders growing major cereals, 10.9 million holders (67%) grew maize in 2016/17 cropping season.

Area coverage of maize in developing countries almost doubled in the last half a century from about 100 million ha in 1965 to more than 190 million ha in 2014 (FAOSTAT, 2015). During the same period, production increased by more than fivefold from around 200 million metric tons (MT) in 1965 to about one billion MT in 2014 (FAOSTAT, 2015), with about 67% of this production coming from low and lower middle income countries (Bekele Shiferaw *et al.*, 2011). Similar trends were observed in Ethiopia. The area coverage of maize in the country doubled in the past 20 years from about 1.1 million ha in 1997/98 to 2.1 million ha in 2016/17 while its production increased from 19.2 million MT to 78.5 million MT during the same period (CSA, 1998; CSA, 2017). The demand for maize in developing countries is still expected to double by 2050 as the recently flourishing rapid economic growth in these countries is driving the crop's growing markets for animal feed and industrial raw material besides its popular utilization as food (Fischer *et al.*, 2011).

Nutritionally, 4.5 billion people in 94 developing countries get at least 30% of their food calories from maize (Bekele Shiferaw *et al.*, 2011). It dominates human diet in the developing countries not only as a source of energy, but also protein and other nutrients. In Africa, where animal protein is scarce and expensive, it accounts for 17 to 60% of the total daily protein supply of individuals in 12 countries (Krivanek *et al.*, 2007) and takes 45% share of protein derived from all staple cereals in eastern and southern Africa (Bekele Shiferaw *et al.*, 2011). In Ethiopia, maize is the dominant source of diet and most important staple as revealed by the fact that it accounted for the highest proportion of the national calorie intake among the other cereals (Guush Berhane *et al.*, 2011) and that about 88% of nationally produced maize is consumed as food (Tsedeke Abate *et al.*, 2015).

Such dependence on maize, however, is one of the causes for one-third of all malnourished children to be found in systems where maize is among the top three crops (Hyman *et al.*, 2008). The apparent reason is the poor quality of protein in maize kernels due to low amount of two essential amino acids, lysine and tryptophan. Consequently, millions of African children and nursing mothers, who depend on maize for their protein requirement, suffered from protein deficiency induced diseases such as stunted growth, weakened immune system and impaired intellectual development (Prasanna *et al.*, 2001).

As mentioned above, high consumption of maize by the human population and well established lysine and tryptophan deficiencies in maize protein motivated the search for a maize kernel with higher concentrations of these essential amino acids in its protein. Fortunately, the persistent efforts of different scientists eventually resulted in the development of new maize varieties, collectively called Quality Protein Maize (QPM), with improved protein quality as well as storage and agronomic qualities similar to conventional maize (CM) (Prasanna *et al.*, 2001).

The requirements for the success of QPM varieties in farmers' fields are no different from that of CM. It has to be adaptable to the farmers' environment with high yield and good agronomic performances. Farmers in developing countries including Ethiopia grow maize in small holder farming systems under rain fed conditions with limited inputs which contribute tremendously for the very low yields in this region (Bekele Shiferaw *et al.*, 2011; Tsedeke Abate *et al.*, 2015). These farming systems are characterized by drought stress, low soil fertility, weeds, pests, diseases, low input availability, low input use and inappropriate seeds (Bekele Shiferaw *et al.*, 2011). Currently, drought is considered the number one threat to maize production in Africa, especially in sub-Saharan Africa where most maize is rainfed (La Rovere *et al.*, 2010; Edmeades, 2013), even more so in Ethiopia where almost all of maize production is rainfed (Tsedeke Abate *et al.*, 2015). Rainfall in this region is very unpredictable in terms of timing (may start very early or very late in the cropping season), quantity (sometimes less than 600 mm/annum) and distribution (high in specific periods of the season and very low at the other times) (Izge and Dugje, 2011). Therefore, improved varieties (QPM or CM) must be proved suitable for drought stressed environments.

For a breeding program aiming to develop high yielding hybrids and synthetic varieties, information about combining ability and heterosis of experimental breeding materials is crucial. Combining ability and heterosis studies provide information on the type of gene action controlling the inheritance of desirable quantitative traits which enable the breeders to define a breeding strategy and to select suitable parents and hybrids for further breeding activities and/or commercialization (Hallauer *et al.*, 2010). Sprague and Tatum (1942) identified two types of combining abilities in which they called the average performance of a line in hybrid combinations as general combining ability (GCA) and the deviation of individual combinations from what is expected on the basis of the average performance of the lines involved as specific combining ability (SCA). Therefore, GCA measures additive gene

actions while SCA is the indication of genes with dominance or epistatic effects. These biometrical tools are able to find out the value of an inbred line through the determination of its ability to combine very well with other lines to produce superior hybrids (Akula *et al.*, 2016).

Along with combining ability, the utilization of heterosis is extremely effective for the genetic improvement of different traits since maize hybrids exhibit heterosis for nearly any trait in nearly every hybrid (Flint-Garcia *et al.*, 2009). Duvick (2001) documented that the tremendous increase in maize yield in the United States between the 1930's and the 1970's was due to the exploitation of heterosis. In twentieth century too, maize breeders have been focusing on developing inbred lines that can produce high yielding hybrids when tested in hybrid combinations (Duvick, 2001; Troyer, 2006). Provision of relevant and basic information on combining ability and heterosis to the breeding community has the capacity to boost up maize production (Ali *et al.*, 2012).

The efficiency of a breeding program is determined not only by combining ability and heterosis but also by additional key parameters such as genotypic and phenotypic variances, heritability, genetic advance and the correlation coefficients of agronomic traits (Nzuve *et al.*, 2014). Phenotypic variation may result from combinations of all the variations occurring in segregating populations of maize attributable to additive genetic effects, non-additive effects due to dominance and interaction of non-allelic genes and environmental effects while genotypic variation refers only to the additive genetic or heritable variation which is responsible for progress resulting from selection (Robinson *et al.*, 1951). The amount of genetic variability for traits under improvement, which is very crucial for the success of any plant breeding program (Sankar *et al.*, 2006), can be detected by the parameters such as genotypic and phenotypic coefficients of variation (Sesay *et al.*, 2016).

Genetic variability can be efficiently exploited by selection depending upon heritability and genetic advance (Bilgin *et al.*, 2010). Heritability has a predictive function in breeding expressing the reliability of phenotype as a guide to its breeding value (Mohsin *et al.*, 2009). It is this breeding value which determines how much of the phenotype would be transmitted to successive generations (Bello *et al.*, 2012). Estimate of heritability helps breeders select for desired traits effectively and achieve maximum genetic gain with little time and resources (Smalley *et al.*, 2004). This heritability estimate when coupled with genetic advance is more reliable and meaningful than the consideration of each parameter (Nwangburuka and Denton, 2012). There is a direct relationship between heritability and genetic advance (Joseph *et al.*, 2015). Genetic advance explains the degree of gain obtained in a character under a particular selection pressure (Ogunniyan and Olakojo, 2014). High genetic advance coupled with high heritability estimates (Bello *et al.*, 2012) and high genotypic coefficient of variation (Nwangburuka *et al.*, 2012) offers the most suitable condition for selection.

Degree of genotypic and phenotypic correlation of traits is also very important because most of economically important traits such as yield are complex in inheritance and may involve several related traits (Robinson *et al.*, 1951). Correlation coefficients show relationships among independent variables and the degree of linear relation between these traits among genetically diverse population for enhanced progress in crop improvement (Hefny, 2011). Practically, correlations are of interest since selection is usually concerned with changing two or more traits simultaneously (Robinson *et al.*, 1951).

To develop high yielding hybrids, significant emphasis should also be paid towards characterization of elite breeding lines and understanding of their genetic diversity and relationships. Molecular markers have contributed extensively to acquire such information. As Prasanna and Hoisington (2003) indicated, accurate assessment of the levels and patterns

of genetic diversity using molecular markers is particularly helpful in maize breeding for (i) maintenance and broadening of genetic base of the elite germplasm; (ii) assignment of lines to heterotic groups; (iii) selection of appropriate parental lines for hybrid combinations; and (iv) generation of segregating progenies with maximum genetic variability for further selection.

There are several molecular markers that are used to characterize maize germplasm and analyze genetic diversity. Inter simple sequence repeats (ISSRs) and single nucleotide polymorphisms (SNPs) are among these markers. ISSR technique is a PCR based technique and mostly a dominant marker system which uses microsatellites as primers in a single primer PCR reaction (Reddy *et al.*, 2002; Vijayan, 2005; Kassa Semagn *et al.*, 2006). This technique is simple, quick and less costly that does not need the use of radioactivity. ISSR overcomes the low reproducibility of random amplified polymorphic DNA (RAPD), high cost of amplified fragment length polymorphism (AFLP) and the prior sequence information requirement of simple sequence repeat (SSR or microsatellite) for crop specific primer synthesis (Reddy *et al.*, 2002; Vijayan, 2005). ISSR markers have extensively been used, among other many applications, for characterization of germplasm and estimation of the extent of genetic diversity at inter- and intra-specific level in a wide range of crop species (Reddy *et al.*, 2002; Vijayan, 2005). SNP is a single base change in a DNA sequence, with a usual alternative of two possible nucleotides at a given position (Vignal *et al.*, 2002; Kassa Semagn *et al.*, 2006). It meets most, if not all, of the criteria of ideal marker system like high polymorphism and even distribution throughout the genome, as well as codominant, accurate and reproducible data provision which can be generated in a high-throughput and cost-effective manner. RFLP and SSR marker systems possess several of these attributes as well. However, they are not truly low cost or highly scalable (Yan *et al.*, 2009). As compared to SSR markers, SNPs are less polymorphic because of their biallelic nature. They easily

compensate this drawback, though, by being abundant, ubiquitous, and amenable to high- and ultra-high-throughput automation (Mammadov *et al.*, 2012). SNPs can be used for a variety of functions in crop improvement, as other genetic markers, including linkage map construction, genetic diversity analysis, marker-trait association and marker-assisted selection (MAS) (Yan *et al.*, 2009).

Therefore, this study was undertaken on advanced quality protein maize inbred lines to study their combining ability and heterosis in their test cross hybrids. Variability of the test-cross hybrids for considered traits was also assessed. Correlation among measured traits, their heritability and genetic advance if 5% of the hybrids were to be selected were also studied. Finally, the inbred lines were characterized at molecular level using ISSR and SNP markers.

2. Literature Review

2.1. Maize overview

Maize, or corn, is a member of the *Maydeae* tribe of the grass family, *Poaceae* (OECD, 2003). There are four species included in the genus *Zea* of which *Zea mays* L. ssp. *mays* is economically important (Doebley, 1983). The other *Zea* spp. referred to as teosintes, are largely wild grasses native to Mexico and Central America (Doebley *et al.*, 1990). Based on archeological records and phylogenetic analysis, it is believed that domestication of maize began about 6,000 to 10,000 years ago from a Mexican wild grass which has been identified as Balsa teosinte, *Zea mays* spp. p*arviglumis* (Smith, 1989; Doebley, 1990; Doebley *et al.*, 1990; Wang *et al.*, 1999; Piperno and Flannery, 2001; Matsuoka *et al.*, 2002; Doebley, 2004). Balsa teosinte was native to the Balsa River Valley on the Pacific slopes of the states of Michoacán and Guerrero, Mexico (Piperno and Flannery, 2001).

Maize is believed to be introduced to Africa arround 1500 AD and spread all over the continent within 500 years (McCann, 2005; Olaniyan, 2015). Accordingly, it reached Ethiopia in 17th century (Huffnagel, 1961) through southern parts of the country (McCann, 2016). Even though maize had been a minor field crop in the country consumed only in the hunger season until the 1980's, it surpassed teff and barley by the mid 1980's, superseded sorghum on low- to mid-altitude fields, replaced coffee in some areas of the south, complemented fields of chat in the southeast and was the primary focus of state farms in the south and west (McCann, 2016). In 2016/2017 meher (main) season, maize was a runner up next to teff in area of production taking 17% out of cereal area (that of teff was 24%) but first in production making up 27% of cereal production followed by teff (17%) (CSA, 2017).

Maize is a diploid crop (2n = 20) with estimated genome size ranging from 2.3 to 2.7 Gb (Arumuganathan and Earle, 1991; Rayburn *et al.*, 1993; Schnable *et al.*, 2009; Zhou *et al.*, 2009). It is estimated that the genome of maize is likely to contain between 42,000 and 56,000 genes (Haberer *et al.*, 2005). It is characterized by a high percentage of repetitive sequences (at least 66%, Haberer *et al.* (2005)) including transposons and retrotransposons (Ananiev *et al.*, 1998; Feschotte *et al.*, 2002; Morgante, 2006; Liu *et al.*, 2007) of which retrotransposons are far more frequent (Haberer *et al.*, 2005). The whole genome of maize is sequenced and a draft genome map of the 2.3 Gb reference genome of maize inbred line B73 is presented by Schnable *et al.* (2009).

Maize is a monoecious annual plant requiring the help of human to disperse its seeds for propagation and survival (OECD, 2003). Since it is a C_4 crop, maize is the most efficient plant for capturing available resources like water, nitrogen and sunlight energy and converting it into food having a great plasticity adapting to extreme and different conditions of humidity, sunlight, altitude, and temperature (Brown, 1999; Brown *et al.*, 2005; Leakey, 2009; Wang *et al.*, 2014).

Maize has a considerable agricultural and economic value as a crop for food for human consumption, feed and fodder for animals and raw material for industrial products such as starch, sweeteners, oil, beverages, glue, industrial alcohol and fuel ethanol (James, 2003; Nuss and Tanumihardjo, 2010; Ranum *et al.*, 2014). Maize, among cereal crops, also presents unparalleled biological attributes as a model organism for fundamental research in genetic diversity and genome evolution (Strable and Scanlon, 2009). Nutritionally, maize contains about 72% starch, 10% protein and 4% fat supplying an energy density of 365 Kcal/100 g (Nuss and Tanumihardjo, 2010).

2.2. Rationale for quality protein maize development

Proteins, the building blocks of life, are made up of 20 amino acids. Although the metabolism of humans and other mono-gastric animals can synthesize the other amino acids, it cannot make nine of them viz. Histidine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Threonine, Tryptophan and Valine. They are known as essential or indispensable amino acids because only our diets can provide them to our body (Food and Nutrition Board, 2001; Vivek *et al.*, 2008). Dietary proteins from animal sources such as meat, poultry, fish, eggs, milk, cheese, and yogurt contain all the nine essential amino acids and are considered complete proteins (Eze, 2011). In contrast, with the exception of soybean, most plant sources of protein such as legumes, nuts, grains, seeds and vegetables tend to have poor amounts of one or more essential amino acids and are considered incomplete proteins (Food and Nutrition Board, 2001; Eze, 2011).

Maize is no exception among cereal grains. It is well known that the quality of its protein is very poor. However, the role of maize for human consumption when expressed in terms of the share of calories from all staple cereals reaches upto 61% in Mesoamerica, 45% in Eastern and Southern Africa and 21% in West and Central Africa and its contribution as a source of protein from all the cereal staples is very similar to its contribution of calories (Bekele Shiferaw *et al.*, 2011). Its grain accounts for about 15 to 56% of the total daily calories in diets of people particularly in Africa and Latin America, where animal protein is scarce and expensive (Prasanna *et al.*, 2001) and it is a primary source of energy supplement in an animal's diet contributing up to 30 percent protein, 60 percent energy and 90 percent starch (Dado, 1999). Hence, millions of African children and nursing mothers suffered from protein deficiency induced diseases (Prasanna *et al.*, 2001).

Protein deficiency has been shown to affect all of the body's organs and many of its systems, including the brain and brain function of infants and young children; the immune system, thus elevating risk of infection; gut mucosal function and permeability, which affects absorption and vulnerability to systemic disease; and kidney function (Prasanna *et al.*, 2001; Otten *et al.*, 2006). The physical signs of protein deficiency include edema, failure to thrive in infants and children, poor musculature, dull skin and thin and fragile hair (Otten *et al.*, 2006). Severe protein malnutrition may cause kwashiorkor (Rolfes *et al.*, 2009). Common symptoms of kwashiorkor include swollen abdomens, listlessness, and hair color changes. Kwashiorkor is sometimes called the "weaning disease" because of the onset of symptoms in many young children at the time of dietary shift from breast milk to soft cereal foods (Lee *et al.*, 2008). Deficiency of essential amino acids in the animal body also results in a number of negative conditions, including reduced appetite, lowered feed intake, lowered body weights, reduced milk and egg production, poor feed efficiency, inferior growth rate and prolonged time to reach maturity (Eze, 2011).

As a result, the persistent efforts of different scientists for a long period of time yielded quality protein maize (QPM) which contains 90% the protein quality of casein milk compared with 40% for CM (Atlin *et al.*, 2011). Consumption of these varieties leads to greater protein utilization and greater rates of growth among malnourished young children (Bressani, 1992).

2.3. The development of QPM

Most of the protein in a mature corn kernel is contained in the endosperm and the germ. Although the endosperm protein is low in quality, it contributes 80% of the total kernel protein as it constitutes the bulk of the grain in contrast to the germ protein which is superior

in quality (Zuber and Helm, 1972; Vasal, 2001b). Therefore, improvements for quality protein maize should target the endosperm.

A CM endosperm consists of a mixture of prolamins (zeins), glutelins, albumins, and globulins which are differentiated by solubility properties. The average proportion of the alcohol soluble zein is 60% followed by the alkali soluble glutelins (34%), both of which are endosperm-specific, while that of the water soluble albumins and the salt soluble glutelins is 3% each (Vasal, 2000; Vasal, 2002). Although all fractions other than zein (called non-zein fractions) are balanced and quite rich in the two essential amino acids – lysine and tryptophan, the high proportion of zein fraction is the primary cause of poor protein quality in maize because zein proteins are low in these amino acids. The germ contains some lysine and tryptophan, which complements the endosperm for improved amino acid balance; however, it is not sufficient for typical maize to be a high-quality protein source (Vasal, 2001b).

The need to improve quality of endosperm maize protein has been recognized for a long time (Osborne and Mendel, 1914). In 1960s, the discovery of high lysine mutant gene *opaque-2* (o_2) (Mertz *et al.*, 1964) and *floury-2* (fl_2) (Nelson *et al.*, 1965) created enthusiasm to improve the quality of protein in maize and resulted in the successive discovery of several other mutants such as *opaque-7*(o_7) (Misra *et al.*, 1972), *opaque-6* (o_6) and *floury-3* (fl_3) (Ma and Nelson, 1975), *mucronate* (*Mc*) (Salamini *et al.*, 1983) and *defective endosperm* (*De-B30*) (Salamini *et al.*, 1997). Two mutants, *opaque7749* and *opaque7455* (o_{11}) (Nelson, 1981) were also identified which have high lysine gene(s) but with still a high level of zein fraction. The specific chromosomal location and the genetic action for some of the mutants are known. The o_2 mutant is located on chromosome-7, fl_2 on chromosome-4, o_7 on chromosome-10, fl_3 on chromosome-8 and *de-B30* on chromosome-7(Babu and Prasanna, 2014). The mutants o_2, o_6, o_7 and o_{11} are completely recessive while the two floury mutants are semi-dominant and

exhibit variable expression for kernel opacity and protein quality depending on the presence of one or more recessives in the triploid endosperm (Babu and Prasanna, 2014). The mutant *De-B30* is dominant and shows dosage effects on kernel opacity and zein content (Soave *et al.*, 1982).

However, of all known mutants, o_2 is the one that has been tried and used extensively in breeding programs around the world and also the use of o_2 gene in combination with other mutants do not offer any better alternative to this gene (Vasal, 2001b). This nutritionally superior maize was therefore named *opaque-2* (o_2) maize, after the o_2 single gene mutation responsible for its improved protein quality as a result of decreased amounts of zein protein (especially α-zein) and increased amounts of albumin, globulin, and glutelin proteins. Despite the nutritional superiority of o_2 maize, it did not become popular with farmers as well as consumers mainly because of negative secondary (pleiotropic) effects of this mutation. These effects were reduced grain yield, chalky and dull kernel appearance, low kernel density, susceptibility to ear rots and stored grain pests, lower rate of germination and greater kernel breakage (Prasanna *et al.*, 2001; Vasal, 2001b; Krivanek *et al.*, 2007; Babu and Prasanna, 2014).

Fortunately, partially hard or vitreous endosperm or "modified" grains had been observed during the process of converting regular corn populations to o_2 versions (Vasal, 2000; Vasal, 2001a; Krivanek *et al.*, 2007). Further investigations identified various endosperm modifier loci called o_2 *modifiers* (Mo_2s) that could favorably alter the grain characteristics, thereby overcoming negative features of the opaque kernel phenotype while maintaining protein quality (Krivanek *et al.*, 2007; Babu and Prasanna, 2014). These modifier loci do not have any effect of their own as such but interact to improve the kernel hardness and appearance and increase kernel weight and density (Sofi *et al.*, 2013). The resulting maize which is

essentially interchangeable with common maize in both cultivation and agronomic characteristics as well as competitive in terms of yield, lodging, disease and pest resistance while retaining the superior lysine and tryptophan content (Vasal, 2001b) is termed by CIMMYT as quality protein maize (QPM). The term QPM now refers to maize homozygous for the o_2 allele, with increased lysine and tryptophan content but without the negative secondary effects of a soft endosperm (Vasal, 2001b). QPM looks and performs like normal maize and can be reliably differentiated only through laboratory tests (Villegas *et al.*, 1992).

QPM essentially has about twice the levels of lysine and tryptophan than normal maize and also increased levels of histidine, arginine, aspartic acid and glycine. It also has reduced levels of glutamic acid, alanine and leucine. The lower levels of leucine especially is an added advantage as it results in a more balanced leucine to isoleucine ratio that helps to liberate more tryptophan (Sofi *et al.*, 2013). As against the mean lysine and tryptophan levels of 2 and 0.4% of total protein in whole grain flour respectively in CM, the corresponding values in QPM are 4 and 0.8% ranging from 2.7 – 4.5% and 0.5 – 1.1% respectively (Krivanek *et al.*, 2007; Vivek *et al.*, 2008; Adefris Teklewold *et al.*, 2015). This is largely due to a decrease in the zein fraction from 47.2% in CM to 22.8% in o_2 mutants (Babu *et al.*, 2013).

2.4. Breeding strategies for QPM development

Research on QPM initially emphasized on development of donor stock by selection for modified grain texture in QPM backgrounds using various selection schemes (Vasal, 2000; Babu and Prasanna, 2014). This was followed by large scale conversion of CM materials with a wide array of genetic background from different agro-climatic zones into QPM using these donor stocks (Villegas *et al.*, 1992; Vasal, 2001a; Krivanek *et al.*, 2007; Sofi *et al.*, 2013; Babu and Prasanna, 2014). Currently, pedigree breeding to develop new inbred lines from

QPM × QPM and QPM × CM crosses has been the focus in addition to conversion of CM genotypes to QPM using backcross conversion method (Krivanek *et al.*, 2007; Babu and Prasanna, 2014).

The breeding of QPM involves manipulation of three distinct genetic systems. The recessive mutant allele of the o_2 gene is the first and central component. The endosperm hardness modifier genes and the amino acid modifiers/genes influencing free amino acid content in the endosperm are the other systems (Krivanek *et al.*, 2007). Consequently, since conventional breeding procedure has been used for QPM breeding, the procedure is highly cumbersome requiring enormous labor, time and financial resources as the o_2 allele has to be in homozygous recessive state along with the polygenic endosperm modifiers. Moreover, it requires selfing, visualization of kernels on light table and rigorous biochemical tests in each backcross generation (Babu *et al.*, 2013).

Fortunately, rapid advances in genome research and molecular technology have led to the use of DNA marker assisted selection (MAS) which holds promise in enhancing selection efficiency and accelerating the process of development of new varieties/hybrids with higher yield potential (Ribaut and Hoisington, 1998). For instance, a rapid line conversion strategy has been developed combining high protein quality and kernel modification in CM inbreds through two-generation backcross program that employs foreground selection for *opaque-2* in both BC_1 and BC_2 generation combined with background selection for recipient genome at BC_2 generation and phenotypic selection for kernel modification and other desirable agronomic traits in two subsequent selfed generations (Babu *et al.*, 2005). While marker-assisted foreground selection (Melchinger, 1990) helps in identifying the gene of interest without extensive phenotypic assays, marker-assisted background selection (Frisch *et al.*, 1999) significantly speeds up the rate of genetic gain/recovery in a backcross breeding

program. There are a few successful examples of MAS for maize improvement using o_2-specific molecular markers (Babu *et al.*, 2005; Gupta *et al.*, 2009; Prasanna *et al.*, 2010; Jompuk *et al.*, 2011).

With the development and access to reliable PCR-based allele-specific markers such as simple sequence repeats (SSRs) and single nucleotide polymorphisms (SNPs), MAS is becoming an attractive option, particularly for oligogenic traits such as QPM (Babu *et al.*, 2004). The cloning and characterization of the o_2 gene, followed by detection of three SSR markers (*phi057*, *phi112* and *umc1066*) within the gene (Yang *et al.*, 2004), led to effective differentiation of the O_2 and o_2 alleles (Bantte and Prasanna, 2003). A combination of bulked segregant analysis (BSA) and genome-wide SNP scan (using Illumina's GoldenGate assay) in phenotypically contrasting progenies have identified several genomic regions putatively associated with kernel hardness and high tryptophan concentration which, if validated using targeted SSRs and segregating populations, will greatly aid in designing a comprehensive MAS system for cost-effective QPM hybrid cultivar development (Babu *et al.*, 2009).

Genetic engineering efforts targeted towards enhancement of lysine content in maize kernels employed RNA interference (RNAi) technology (Huang *et al.*, 2006; Wu and Messing, 2011) and deregulation of the aspartate metabolic pathway (Wang *et al.*, 2007; Frizzi *et al.*, 2008).

2.5. Benefits of QPM

Small holder farmers in Africa plant maize as both a household garden food and cultivated field grain (McCann, 2005). A small percentage of the crop is eaten as fresh maize, which is boiled or roasted as a snack; kernels are stripped from the cob, dried, and then stored or sold for further processing (Nuss and Tanumihardjo, 2011). The vast majority of maize is harvested at maturity, and the dried kernels are often milled into flour or grits using various

wet or dry techniques (Nuss and Tanumihardjo, 2010). Such diverse and heavy consumption of maize renders QPM prominent value in maize-sustaining households. Bressani (1992) showed that increased concentration of lysine and tryptophan in the grain endosperm of QPM can double the biological value of maize protein. In other words, the amount of CM that needs to be consumed to achieve amino acid equilibrium is more than twice as much as the amount of QPM (Nuss and Tanumihardjo, 2011). The nutritive value of milk protein is considered to be higher than that of maize protein; however, milk is a protein source that very few people can afford. QPM protein is more bioavailable (National Research Council, 1988) and it has a quality value equivalent to 90% that of milk (FAO, 1992). In Colombia, children suffering from kwashiorkor were restored to normal health with a diet containing only QPM as the protein source (Vivek *et al.*, 2008). Therefore, the major benefit of QPM for human nutrition is found in decreasing deficiency of amino acids (Krivanek *et al.*, 2007). Vivek *et al.* (2008) reported that studies had shown the support of increased levels of lysine in the assimilation of zinc and iron from maize grain which can be considered as an added benefit of QPM.

Another application of QPM is as animal feed, especially for monogastric animals such as pigs and poultry, which require a more complete protein than cereals alone can provide, as is the case with CM that is deficient in lysine and tryptophan. A number of studies have proved that the more potential impact of QPM can be its use in commercial feeds for pigs and poultry as it results in improved growth (Sofi *et al.*, 2013). Pigs raised on high lysine/tryptophan maize gain weight at roughly twice the rate of animals fed solely on CM with no additional protein supplements (Vivek *et al.*, 2008). Performance of finishing swine was greater for those fed on QPM than for those fed on CM (Dado, 1999). Qi *et al.* (2002) showed that the energy available from QPM is little higher than that from CM. The authors also observed that QPM not only had a higher content of lysine but it was also digested better

by pigs due to the improvement of protein quality by a higher level of albumins/globulins. According to the authors, CM contributes up to a third or more of the crude protein content of chicken diets and thus, feeding CM necessitates the use of expensive protein ingredients, including fishmeal and soybean meal. They also reported that nutritional evaluation of QPM in various locations has proved the superiority of QPM over CM in the feeding of chicken.

2.6. Combining ability

Combining ability is an especially powerful tool for studying and comparing the performance of inbred lines in hybrid combinations (Griffing, 1956). Information on the combining ability of maize germplasm is of great value to maize breeders. Combining ability of inbred lines is the ultimate factor determining future usefulness of the lines for hybrid development (Hallauer *et al.*, 2010). It is the potential of an inbred line to fit in crosses with a number of inbred lines or any one of inbred line for revealing desirable attributes in hybrid combinations (Hallauer *et al.*, 2010). Combining ability has also been described as the ability of a parent to produce inferior or superior combinations in one or a series of crosses (Chaudhary, 1982). Combining ability of inbred lines is the ultimate factor determining future usefulness of the lines for hybrid development (Hallauer *et al.*, 2010).

Several mating designs could be employed to study the combining ability of parental inbred lines. Among them, the diallel mating design is used extensively (Griffing, 1956). However, this mating design, where $n(n-1)/2$ combinations are developed from a set of n inbred lines, is possible with relatively few inbred lines. When the number of inbred lines is too many to manage, the testcross test introduced by Davis (1927) is the best option (Hallauer *et al.*, 2010). Line × tester mating design was developed by Kempthorne (1957), which provides reliable information on the general and specific combining ability effects of parents and their hybrid combinations in applied breeding programs. The design has been widely used in maize

breeding by several workers and continues to be applied in quantitative genetic studies in maize due to its significance (Abuali, 2016).

The relative importance of additive (GCA) versus non-additive (SCA) effects is an indication of the type of gene action (Baker, 1978). The proportion of additive and non-additive components of genetic variance depends on the genetic structure of the crosses analyzed and the environmental conditions in which they were grown (Khotyleva and Trutina, 1973). In previously unselected materials, additive gene action is more important than non-additive components for most traits (Younes and Andrew, 1978). In materials that were previously selected for GCA, however, non-additive gene action was more important for some traits, including grain yield (Kambal and Webster, 1965).

Several authors have examined the combining ability of different maize genotypes of various genetic structures using line × tester mating designs and tested them under different environmental conditions to determine their general and specific combining abilities. Legesse Welde *et al.* (2009) used factorial cross (Design II) of 26 inbred lines with 6 testers and evaluated the resulting 156 F_1s along with 2 checks in 5 mid-altitude and highland areas of Ethiopia. They found highly significant GCA and SCA mean squares due to lines and testers for most studied traits while GCA/SCA mean squares exhibited the preponderance of additive gene effects.

In line × tester analysis of elite maize inbred lines at Melkassa, Shushay Welderufael *et al.* (2013) observed significant mean squares due to GCA of lines, and testers and SCA of line × tester for grain yield and most yield related traits. The authors reported the importance of both additive and non additive gene actions in controlling those traits.

Tessema Tamirat *et al.* (2014) also studied combining ability of 36 maize inbred lines crossed with two testers at Melkassa and they found significant GCA mean squares but non-significant SCA mean squares for all traits demonstrating that variation among crosses was due to additive gene effect.

Amare Seyoum *et al.* (2016) evaluated 34 highland maize testcrosses for 17 traits at Jimma, south west Ethiopia and reported highly significant GCA mean squares due to lines for most traits while SCA mean squares were significant only for some of the traits. The authors also reported higher percentage for relative contribution of GCA over that of SCA in all studied traits indicating the predominance of additive gene effect in controlling the inheritance of those traits.

Gudeta Nepir *et al.* (2015) evaluated hybrids of 20 QPM inbred lines and two testers across three highland areas of Ethiopia and found that mean squares attributable to GCA and SCA effects were significant for most traits along with higher contributions of GCA sum of squares to the variation among the hybrids than SCA sum of squares suggesting that the traits were conditioned mainly by additive gene effects.

2.7. Heterosis

The term heterosis is more widely associated with maize than any other important crop species (Hallauer and Carena, 2009). The decrease in vigor and yield (inbreeding depression) that follows inbreeding and the increased vigor (heterosis) that so frequently follows crossing have long been recognized in maize (Richey, 1927). Heterosis or hybrid vigor and inbreeding depression are complementary. Maize breeding methods have been developed to take advantage of the manifestation of heterosis in crosses of inbred lines (Hallauer *et al.*, 2010). Heterosis can be defined as the hybrid vigor manifested in hybrids and represents the

superiority in performance of hybrid individuals compared with their parents (Hallauer *et al.*, 2010).

Although heterosis has been exploited extensively in breeding and commercially, it is poorly understood. Despite extensive study, its genetic basis remains unresolved (Troyer, 2006; Hallauer, 2007). Theories advanced to explain the phenomenon include interaction of dominant favorable alleles, intra-allelic interactions (overdominance), inter-allelic interactions (epistasis), complementary action of linked genes (pseudo-overdominance), the non-genetic physiologic stimulation (Hallauer and Carena, 2009), dispersion of completely or incompletely dominant genes and unidirectional dominance (Mather and Jinks, 1982).

Heterosis can be inferred from heterotic patterns (Hallauer and Carena, 2009). A heterotic pattern can be defined as the cross between known genotypes that expresses a high level of heterosis (Carena and Hallauer., 2001). They had been established empirically by relating the heterosis of crosses with the origin of the parents included in the crosses based on diallel crosses studies on performance (Hallauer *et al.*, 1988). Due to different earlier works by different scientists, it had been a common knowledge that hybrids of lines from different germplasm sources or crosses of distantly related parents had greater yields than hybrids of lines from similar sources or crosses of more closely related parents indicating genetic divergence of parental crosses was important for expression of heterosis (Hallauer *et al.*, 2010). However, Moll *et al.* (1965) concluded that heterosis increased with increased divergence within a restricted range of divergence as extremely divergent crosses resulted in a decrease in heterosis.

The identification of heterotic patterns is a consequence of time, experience and effort to acquire good germplasm knowledge and to conduct extensive testing. Biochemical or molecular markers have been very useful to assess genetic diversity and genetic divergence.

However, they have limited usefulness for predicting good heterotic combinations. Consequently, Melchinger (1999) concluded that evaluating the performance of crosses among groups based upon genetically diverse parents is essential to identify promising heterotic patterns. Barata and Carena (2006) also noticed inconsistency between genotypic and phenotypic (e.g., testcross, diallel) data which were attributed to the complexity encountered in multi-trait and multi-stage selection for economically important traits and they recommended extensive testing of phenotypic data to be a priority of maize breeding programs.

Many studies have been undertaken to determine heterosis using different maize genotypes from different sources at different locations. Rodrigues *et al.* (2006) detected significant average heterosis in grain yield, number of days to female flowering and to all evaluated diseases in partial diallel crosses of 16 white grain maize populations presenting high quality protein. From the result, they concluded the importance of dominance gene in controlling grain yield. In a diallel cross of eight inbred lines, Dagne Wegary *et al.* (2007) found mean mid-parent heterosis (MPH) ranging from 2.9% for days to maturity to 89.2% for grain yield and high-parent heterosis (HPH) ranging from 0.65% for ear diameter to 64% for grain yield. The authors also observed that all the crosses exhibited positive MPH for ear and plant height, ear length, kernels per row and grain yield. Makumbi *et al.* (2011) reported the highest MPH for GY that ranged from 74% to 1119% under drought stressed environments as compared to the MPH ranging from 14% to 448% under well-watered environments. Furthermore, they reported that MPH was significantly correlated with GY and SCA under drought, low N stress and well-watered conditions.

Several studies have been conducted in Ethiopia to determine heterosis in different maize genotypes. Shushay Welderufael (2014) noticed substantial standard heterosis for various

traits in line × tester crosses that involved 24 lines and 2 testers. The author reported the highest standard heterosis of 89.24% over the popular QPM hybrid (BHQPY545) and 131.20% over the most popular drought tolerant open pollinated variety (Melkassa2). He inferred that this result indicated the presence of substantial heterotic potential that could be exploited in maize breeding program and the possibility of developing desirable cross combinations and synthetic varieties through crossing and/or recombination of inbred lines with desirable traits of interest. Using the North Carolina Design II mating scheme, Wende Abera *et al.* (2016) found maize hybrids that had up to 235% and 250% high-parent and mid-parent heterosis for grain yield and up to –26% mid-parent heterosis for northern corn leaf blight (NCLB) reaction.

2.8. Molecular markers

Integration of molecular tools in maize breeding programs to enhance breeding efficiency and effectiveness has become fundamental to many national agricultural research systems (NARS) worldwide. There are several molecular markers used for plant genetic analysis. The potential of DNA markers to aid plant breeding programs is diverse that include fingerprinting of elite genetic stocks, analysis of genetic diversity and increasing the efficiency of selection for difficult traits (Prasanna and Hoisington, 2003). Among so many DNA based markers available to plant scientists, the ones most commonly used are restriction fragment length polymorphisms (RFLPs), random amplified polymorphic DNAs (RAPDs), amplified fragment length polymorphisms (AFLPs), simple sequence repeats (SSRs or microsatellites), inter-simple sequence repeats (ISSRs) and single nucleotide polymorphisms (SNPs). Each one of these marker systems offers a unique combination of advantages and disadvantages (Kumar et al., 2009). They differ in the type of sequence polymorphism detected, the information content, the dominance relationships between alleles, the amount of

DNA required, the need for DNA sequence information in the species under analysis, the development costs, the ease of use and the extent to which they can be automated (Kumar et al., 2009). The choice of marker systems is to significant extent dictated by the specific application and there is not a single class of markers that can satisfy all the needs encountered by plant geneticists and breeders. The two DNA based markers that were applied in the current study i.e. ISSR and SNP markers are reviewed in the next sub-sections.

2.8.1. Inter-simple sequence repeat (ISSR) marker

ISSR is a novel PCR-based technique that amplifies DNA sequences of about 100–3000 bp located between adjacent, oppositely oriented (inverted) and identical microsatellite repeat regions (Zietkiewicz *et al.*, 1994; Kumar *et al.*, 2009). ISSRs are amplified by PCR using usually 16–25 bp long microsatellites as primers in a single primer PCR reaction targeting multiple genomic loci (Reddy *et al.*, 2002; Kassa Semagn *et al.*, 2006). The primers used can be either unanchored (Gupta *et al.*, 1994; Wu *et al.*, 1994) or more usually anchored at 3' or 5' end with 1 to 4 degenerate bases extended into the flanking sequences (Zietkiewicz *et al.*, 1994). While unanchored primers form smears due to slippage within the repeat units during amplification, more number of bands and a higher degree of polymorphisms are observed when 5' anchors are used (Reddy *et al.*, 2002). However, 3' anchored primers give clearer banding pattern as compared to those anchored at 5' end (Nagaoka and Ogihara, 1997). ISSRs can be detected using polyacrylamide gel electrophoresis (PAGE) in combination with radioactivity or with silver ($AgNO_3$) staining and agarose gel with ethidium bromide (EtBr). The former is the most sensitive while the latter is the least sensitive. Reddy *et al.* (2002) explained in depth that failure in mismatch repair during DNA replication, mutation at the priming site, the absence/presence of anchorage and the sequence of the repeats (motifs) in the primers and the detection methods are the sources of the variability in ISSRs.

Even though ISSRs have higher reproducibility than RAPDs due to the use of longer primers (16–25 mers) and thereby higher annealing temperature (45–60°C) giving them higher stringency, reproducibility is often stated as one of the limitations of ISSRs. However, Moreno *et al.* (1998) reported about 92–95% repeatability across DNA samples of the same cultivar and across PCR runs when detected using PAGE. The other limitation of ISSRs is their segregation mostly as dominant markers (Gupta *et al.*, 1994; Wang *et al.*, 1998) although rarely as co-dominant (Wu *et al.*, 1994; Akagi *et al.*, 1996; Wang *et al.*, 1998; Sankar and Moore, 2001).

ISSRs were successfully used by various investigators to assess the genetic diversity of maize. Kantety *et al.* (1995) investigated the potential of ISSR markers to detect the diversity among nineteen popcorn and eight dent corn inbred lines using ten primers and reported that ISSRs appear to correspond to known pedigree information and can be used for classification of inbred lines in hybrid testing. They also found that ISSRs can be used to identify polymorphic loci among closely related lines. Carvalho *et al.* (2002) examined the genetic diversity among 79 landraces and two improved varieties of maize in Brazil with nine ISSR primers and the results revealed that ISSR markers could be efficiently used to quickly access the genetic variation available in the maize germplasm. Abdellatif and Khidr (2010) studied genetic diversity of four new yellow single crosses, five new yellow three-way crosses, and five yellow inbred lines of maize using molecular markers RAPD, ISSR, SSR and biochemical markers. Their results indicated that all the three molecular markers are efficient for evaluating genetic diversity in the maize genotypes with moderately high correlations between ISSR and SSR (0.67) and ISSR and RAPD (0.65). do Amaral Júnior *et al.* (2011) analyzed the genetic diversity among 52 different maize types (pop, common, tetraploid and sweet) using 15 ISSR primers and the efficiency of ISSR molecular markers in the separation and identification of the variability of maize accessions was demonstrated by the resulting

dendrogram. Nkongolo *et al.* (2011) assessed the level of genetic variation and relatedness among and within QPM and normal maize varieties using ISSR and RAPD markers and higher genetic variations among accessions was observed with RAPD compared to ISSR primers. However, both the markers exhibited that no significant differences among the QPM and normal maize varieties and the different maize accessions were closely related genetically. Mbuya *et al.* (2012) assessed the genetic diversity among 137 maize inbred lines using four ISSR primers and found a high level of variability among and within the inbreds. They concluded that ISSR data are useful in the selection of candidate inbreds for testing for general and specific combining abilities. Idris *et al.* (2012) evaluated the genetic diversity among nine maize inbred lines using eight ISSR primers and concluded that ISSR markers could be used as a reliable method for the characterization of maize genotypes. Žiarovská *et al.* (2013) successfully discriminated Iowa Stiff Stalk Synthetic and Iodent Reid populations of maize by analyzing genetic diversity using 12 ISSR primers. Lenka *et al.* (2015) assessed 49 maize inbreds using 12 ISSR primers. They identified highly divergent lines which could be sorted out as valuable materials for heterosis breeding for production of single cross hybrids.

Nkongolo *et al.* (2015) successfully identified variety diagnostic marker for one of the varieties in their study using 24 ISSR and 46 RAPD primers which indicates the usefulness of ISSR markers to track varieties in progenies in targeted breeding programs. Detail description of applications of ISSR markers in genomic fingerprinting, phylogeny, gene tagging and marker assisted selection, genome mapping, determining SSR motif frequency and evolutionary biology in different crops is provided in Reddy *et al.* (2002).

2.8.2. Single nucleotide polymorphism (SNP) marker

SNPs are simply loci in the genome where the sequence of DNA differs by a single base with a usual alternative of two possible nucleotides at a given position. This largely biallelic nature of SNPs is attributable to the occurrence of only two of the theoretically possible four nucleotides in reality at a particular site in a population (Jehan and Lakhanpaul, 2006). Such a base position can be considered as SNP only if different sequence alternatives (alleles) exist at that position in normal individuals in certain population(s) and if the rarest allele has an abundance of 1% or greater in the population(s) (Brookes, 1999). This description separates single base insertion or deletion variants (indels) from SNPs. SNPs were proved to be universal and the most abundant forms of genetic variation among individuals of the same species (Rafalski, 2002). Using US elite maize germplasm, Bhattramakki *et al.* (2002) discovered 1 SNP per 48 bp and 131 bp on average in non-coding and coding regions, respectively.

Development of SNP markers usually involves SNP discovery and SNP validation in order to use them for genotyping. Genome-wide detection of SNPs in complex genomes has become achievable due to the availability of reference sequences, the application of genome complexity reduction techniques and NGS technologies coupled with post-re-sequencing computational treatment which are the prerequisites (Kumpatla *et al.*, 2012). Once true SNPs have been discovered, they have to be validated to be converted into a valid marker. The validation of a marker is a process of designing an assay based on the discovered polymorphism and genotyping a panel of diverse germplasm or segregating population (Kumpatla *et al.*, 2012). The most popular high throughput assays, chemistries and genotyping platforms that are currently being used for SNP validation are Illumina's BeadArray technology-based Golden Gate (GG) (Fan *et al.*, 2003) and Infinium assays

(Steemers and Gunderson, 2007), Life Technologies' TaqMan (Livak *et al.*, 1995) assay coupled with OpenArray platform (TaqMan OpenArray Genotyping system, Product bulletin, 2010) and KBiosciences' Competitive Allele Specific PCR (KASPar/KASP) complemented with the SNP Line platform (SNP Line XL; http://www.kbioscience.co.uk).

SNP markers have been successfully used for characterization of different germplasm. Lu *et al.* (2009) characterized a total of 770 maize inbred lines including 394 lines from CIMMYT-Mexico, CIMMYT-Zimbabwe and CIMMYT Kenya, 282 lines from China and 94 lines from Brazil with 1,034 SNP markers. Pairwise comparison between these three sets showed that the temperate (Brazilian and Chinese) and tropical/subtropical (CIMMYT) germplasm clearly clustered into two separate groups and the temperate germplasm could be further divided into six groups consistent with known heterotic patterns in contrast to the tropical/subtropical germplasm. Their result also showed that all the different genotypes utilized limited genetic diversity existing in the center of origin. They finally recommended a subset of 449 (out of 1034) high quality markers for routine breeding activities.

Wen *et al.* (2011) assembled 359 maize inbred lines from 12 different breeding programs representing the genetic diversity of CIMMYT and IITA's breeding programs for drought, acid soils, low N, disease and insect resistance to be genotyped using 1260 SNP markers. Multivariate analyses performed in the study showed that the lines related by pedigree tended to cluster together. They were in line with nine main subsets of lines determined in the study based on pedigree information, environmental adaptation and breeding scheme. The analysis of molecular variance (AMOVA) revealed that variation within these subsets was much higher than that among subsets. This study, unlike other studies, additionally evaluated various models for association analysis based on this panel of inbreds and concluded that the panel is ideal for association mapping.

Kassa Semagn *et al.* (2012) genotyped 450 maize inbred lines developed and/or widely used by CIMMYT-Kenya and CIMMYT-Zimbabwe using 1065 SNP markers and found the uniqueness of the majority of the inbred lines in these maize breeding programs. Different multivariate analyses performed showed that there were three distinct groups which generally agree with pedigree information. However, the SNP markers did not show clear separation of CIMMYT's heterotic groups A and B that were established based on combining ability tests. The authors finally recommended 644 of the 1065 SNPs for low to medium density genotyping in tropical maize germplasm using uniplex assays.

Dao *et al.* (2014) characterized a total of 100 maize inbred lines including 59 lines from Burkina Faso (INERA), 16 lines from CIMMYT-Zimbabwe, 15 lines from IITA and 10 temperate lines from France using 1057 SNP markers. The multivariate analyses executed revealed five groups that somewhat agree with the origin of the germplasm. Pairwise comparisons between INERA and the other germplasms showed that the temperate and some IITA lines were differentiated from INERA lines. In this study, 580 high quality SNPs out of 1057 were identified and recommended.

Wu *et al.* (2016) used a total of 538 CIMMYT maize inbred lines (CMLs) including 283 lowland tropical lines from Mexico, Kenya, Zimbabwe and Thailand, 225 subtropical/mid altitude lines from Mexico, Kenya, Zimbabwe and Colombia, 30 highland tropical lines from Mexico and 6 temperate inbred lines for molecular characterization analysis. Selected 362,008 SNPs were used for the analysis in the study. Their results indicated the uniqueness of most lines in the current CML collection and clear population structure and divergence between the temperate and tropical groups of inbreds. In line with former authors, these scientists also confirmed that the heterotic patterns in tropical maize collections are not clear compared with temperate maize, and the heterotic patterns estimated based on molecular

markers are not fully consistent with those estimated based on combining ability tests and pedigree information.

In addition to the above mentioned use in diversity studies, SNPs have been effectively applied in QTL discovery (Zheng *et al.*, 2008; Buckler *et al.*, 2009; Poland *et al.*, 2011), genome wide association study (GWAS) (Weng *et al.*, 2011) high density genetic mapping (Wang *et al.*, 2015) and plant breeding including different marker assisted selection (MAS) approaches like marker assisted back crossing (MABC) (Ribaut and Ragot, 2007), marker assisted recurrent selection (MARS) (Massman *et al.*, 2013; Yoseph Beyene *et al.*, 2016) and genomic selection (GS) (Yoseph Beyene *et al.*, 2015). SNP markers associated with phenotypic changes identify the largest class of functional polymorphism.

2.9. Maize genetic analysis and QPM research in Ethiopia

QPM research in Ethiopia was started in 1980 by testing introduced QPM pools and populations from CIMMYT (Leta Tulu *et al.*, 2002). The authors also documented that introduction has continued since then and conversion of locally adapted varieties to QPM started in 1990's. In addition, they recorded that hybrid development was initiated by introduction of lines and hybrids annually from CIMMYT and other national agricultural research centers. The first QPM variety released in Ethiopia was BHQP542 which was released in 2002 (Leta Tulu *et al.*, 2002; National Agricultural Input Authority, 2003). Since then, three open pollinated varieties (OPVs) namely Melkassa6Q (2008), Melkassa1Q (2013) and Gibe3 (2015) and four hybrids namely BHQPY545 (2008), AMH760Q (2012), MHQ138 (2012), and BHQP548 (2015) were released by Ethiopian Institute of Agricultural Research (EIAR) (MoANR, 2016).

Several authors in Ethiopia have carried out genetic analysis studies on both QPM and CM. However, most of the combining ability and heterosis analysis of maize in Ethiopia were focused on mid-altitude and highland sub-humid genotypes (Dagne Wegary *et al.*, 2007; Dagne Wegary, 2008; Legesse Welde *et al.*, 2009; Dagne Wegary *et al.*, 2014; Girma C. Hosana *et al.*, 2015; Gudeta Nepir *et al.*, 2015; Amare Seyoum *et al.*, 2016; Demissew Abakemal *et al.*, 2016; Wende Abera *et al.*, 2016). There have been few analyses using genotypes developed for drought stressed areas (Shushay Welderufael *et al.*, 2013; Shushay Welderufael, 2014; Tessema Tamirat *et al.*, 2014; Mieso Keweti *et al.*, 2016). However, they used only CM inbred lines in their experiments. In addition, no researcher has ever carried out diversity analysis on genotypes developed for drought stressed areas using molecular markers, let alone ISSRs and SNPs.

Moreover, continuous improvement of maize is very important for the increased productivity of the crop (Ogunniyan and Olakojo, 2014). This can be achieved through effective selection of suitable parental materials of significant genetic variability using different parameters. Therefore, since the genotypes in this study were newly developed which have not been analyzed, they should be evaluated for their per se performance as well as genetic analysis to select best inbred lines for further breeding and hybrid development. Hence, this study was initiated with the following objectives.

3. Objectives

3.1. General objective

To determine genetic parameters of elite quality protein maize inbred lines for grain yield and other agronomic traits in drought stress environments and investigate their genetic diversity and relationship using SNP and ISSR markers.

3.2. Specific objectives

1. To estimate general and specific combining ability effects for grain yield and other agronomic traits of QPM inbred lines
2. To estimate the potential of QPM inbred lines to produce heterotic hybrids (heterosis) for grain yield and other agronomic traits;
3. To estimate the genetic variation, trait correlation, heritability and expected genetic advance in QPM inbred lines
4. To examine genetic differentiation, population structure and patterns of relationship among the inbred lines using SNPs and ISSR markers

4. Materials and Methods

4.1. Germplasm

Fifty eight fixed (S7) early maturing QPM inbred lines derived through backcross breeding method and two widely used QPM inbred line testers, CML144 and CML159 (Appendix 1) were used for this study. The lines were introduced from CIMMYT-Kenya at S_2 stage of inbreeding generation and advanced up to S_7 stage at MARC. They were developed from QPM versions of eight synthetic varieties, namely, ECA-EE-6, ECA-EE-8, ECA-EE-9, ECA-EE-13, ECAEE-16, ECA-EE-31, ECA-EE-33 and ECA-EE-34. A QPM pool called $PL15QPMC_7SRC_1F_2$ was used as a recurrent parent and crossed to the synthetics. POOL15QPMSR is one of the first seven tropical QPM gene pools formed by CIMMYT with resistance to maize streak virus. This pool was reported to have 9.1% protein content and 0.94% and 4.2% tryptophan and lysine content in grain protein, respectively (Prasanna *et al.*, 2001; Vasal, 2002). The resulting single crosses were then backcrossed to POOL15QPMSR (the recurrent parent) to form BC_1F_1. The BC_1F_1 was then selfed and ears bulked to form S_1 bulks. These bulks were advanced up to S_7 by selfing and shelling ears individually during the subsequent seasons except at S_6 where all harvested ears in a row were bulked. At each generation, selected best ears were planted using ear to row method in a separate selfing block. Simultaneously, rigorous field selection was made based on per se performance of the lines at Melkassa Agricultural Research Center (MARC) whereby only inbred lines or individual plants selected for desirable agronomic performance, disease (especially rust, which is the most important disease in drought stressed maize growing areas of Ethiopia) tolerance and early maturity were selected in the field and advanced to the next generation of inbreeding. After harvesting, those lines with poor ear aspect (kernel texture, kernel row arrangement, grain filling, husk cover etc) were discarded. Through the process of line development, four lines were selected from ECA-EE-6 background (hereafter called

population A), thirteen lines from ECA-EE-8 background (hereafter called population B), four lines from ECA-EE-9 background (hereafter called population C), eight lines from ECA-EE-13 background (hereafter called population D), eight lines from ECA-EE-16 background (hereafter called population E), one line from ECA-EE-31 background and two lines from ECA-EE-33 background (hereafter called population F) and eighteen lines from ECA-EE-34 backgrounds (hereafter called population G) with two QPM testers (hereafter called CMLs or population H) were used in this study. Designation, pedigree and tryptophan content of the 58 inbred lines used for this study were presented in Appendix 1.

The 58 inbred lines were then crossed in a line by tester (L×T) mating design during 2014 off-season (from January to May) at MARC under irrigation and a total of 116 test crosses were generated. The parental lines were planted in two rows of 5 m length, whereas 15 rows of the same length were used to plant the testers on three planting dates separated by 7 days interval. Pollen from the plants of one tester was used to cross each plant in one of the rows of the female lines and pollen from the plants of the other tester was used to pollinate each plant of the other row of the female lines. Seed increase of the lines was also done simultaneously using plant to plant sibbing (full sib) method on a 3 m row for each line.

4.2. Field experiment

4.2.1. Experimental sites

This study was conducted at three sites (MARC, Edo Gojola and Mieso) that represent mid-altitude drought stressed areas of Ethiopia. MARC is drought stress tolerant maize research project coordination center of EIAR. It is located at 8°24'N latitude and 39°12'E longitude, 115 km South East of the Ethiopian capital city – Addis Ababa. The center is situated at 1540 meters above sea level (masl). It receives 734 mm average annual rainfall and has mean minimum and maximum temperature of 14.1°C and 28.4°C, respectively. In addition to MARC, Edo Gojola research center (formerly Ziway testing site) under Hawassa University and Mieso research center were used for this study as they can represent Southern and Eastern Ethiopia low moisture stressed regions. Edo Gojola research center is 160 km away to the South of Addis Ababa situated at 7°56'N latitude and 38°35'E longitude. It is located near Ziway town at an elevation of 1640 masl. The mean minimum and maximum temperature of the center are 13.7°C and 26.7°C, respectively and receives 760 mm of mean annual rainfall. Mieso research center is located about 300 km East of Addis Ababa at 8°48'N latitude and 40°9'E longitude at an altitude of 1327 masl with an average annual rainfall of 801 mm. The center has mean minimum and maximum temperature of 14.6°C and 30.3°C, respectively.

4.2.2. Experimental designs

The 116 test cross F_1 hybrids developed by crossing 58 inbred lines with two testers and the 58 inbred lines were evaluated in two separate sets of trials laid out in contiguous plots. The experiments were planted across three drought stressed areas – MARC, Edo Gojola and Mieso in 2014 main season. One released QPM hybrid from MARC, MHQ138, one yellow

QPM hybrid released from Bako National Maize Research Center (BNMRC), BHQPY545, and two CM hybrids released from MARC, MH140 and MH130, were used as standard checks for the hybrid trials. CML144 and CML159, CIMMYT QPM lines commonly used across Eastern and Southern Africa (ESA) region were used as checks for the inbred line trials. The experimental design used for both trials was alpha (0,1) lattice (Patterson and Williams, 1976) with 10 plots (each plot consisting of 2 rows) of 4 m length and 12 incomplete blocks (10 × 12) for the 120 hybrids (116 F_1 hybrids plus four checks) trial. For the inbred line trial that consisted of 58 S_7 lines and two checks, 12 incomplete blocks of 5 plots each (5 × 12) with two rows of 4 m length per plot were used. Both trial sets were replicated twice at all sites.

4.2.3. Field management

At planting, two seeds were sown per hill with spacing of 25 cm between hills (intra-row spacing) and 75 cm between rows (inter-row spacing) as recommended for drought stressed areas of Ethiopia. They were later thinned to one plant per hill to get a final plant population of 53,333 plants per hectare. Fertilizer application was also done as recommended for the area by applying 46 kg P_2O_5 per hectare as diammonium phosphate (DAP) at planting and by top dressing 41 kg Nitrogen (N) per hectare as urea by side dressing at knee height stage (30-35 days after emergence). The field was kept as free as possible from any weed throughout the experimentation by hand weeding.

4.2.4. Data collection and measurements

Data were recorded on 18 different quantitative characters on each plot. List of all the collected data and the way in which measurements were recorded or calculated are presented in Table 1.

Table 1. List of data collected from different traits, their abbreviation, units and description

No	Abbreviation	Trait	Unit	Trait description
1	DA	Days to anthesis	days	Number of days from planting to when 50% of the plants in a plot shed pollen
2	DS	Days to silking	days	Number of days from planting to when silks of 50% of the plants in a plot emerge
3	ASI	Anthesis silking interval	days	Calculated as the difference between DA and DS (ASI = DS – DA)
4	CLR	Common leaf rust (*Puccinia sorghi*)	score (1–5)	Scored on a scale of 1 to 5 taking 1 as free from rust symptom and 5 as severely infected
5	PH	Plant height	centimeters (cm)	The height from ground level to the insertion of the first tassel branch measured from five randomly selected plants before the plants started to dry out; average of the five plants were used to record a plot data
6	EH	Ear height	cm	The height of the same five plants measured from ground level to the node at which the top ear is developed before the plants start to dry out and averaged
7	RL	Percentage of lodged plants at root level	percent (%)	Percentage of plants leaning more than 45° from the vertical
8	SL	Percentage of lodged plants at stalk level	percent (%)	Percentage of plants broken at or below the node of top ear
9	EPP	Number of ears per plant	number	Calculated by dividing the number of ears that have at least one fully developed grain to the number of plants at harvest from which those ears are harvested

Table 1. List of data collected continued

No	Abbreviation	Trait	Unit	Trait description
10	PA	Plant aspect	scale (1–5)	The overall architecture or appeal of the plants of each plot on a scale of 1 to 5 taking 1 as having the most attractive plants
11	EA	Ear aspect	scale (1–5)	The overall appearance of the ears of each plot on a scale of 1 to 5 taking 1 as having the most attractive ears
12	EL	Ear length	cm	The length of five representative ears from each experimental plot measured from their base to tip using digital caliper and averaged
13	ED	Ear diameter	cm	The diameter of the same five ears measured at the mid-section along the ear length using digital caliper and averaged
14	RPE	Rows per ear	number	Number of kernel rows around the perimeter of the cob on the same five ears and averaged
15	KPR	Kernels per row	number	Number of kernels on one full length row measured from the same five ears and averaged
16	TKW	Thousand kernel weight	grams (g)	The weight of 1000 kernels that is estimated from a sample weight of 200 kernels adjusted to 12.5% moisture content
17	GY	Grain yield	tons per hectare ($t\ ha^{-1}$)	Calculated from shelled grain weight or field weight of all the ears from each plot taking shelling percentage of 80% (800 g grain per kg ear weight) and adjusting it to 12.5% moisture content

4.3. Laboratory experiment

4.3.1. Tryptophan analysis

Since tryptophan can be used as a single parameter for the evaluation of the nutritional quality of maize protein due to its strong positive relationship with lysine in the protein of QPM endosperm (Villegas et al., 1984), tryptophan content in whole grain of all the 58 inbred lines was determined at CIMMYT's Cereal Quality Laboratory in Mexico following procedures described by Villegas et al. (1984). The kernels of each of the lines were screened under a light box and rated on a scale of 1–5, with 1 indicating 100% vitreous (with no opaqueness), 2 indicating 25% opaque (75% modification), 3 indicating 50% opaque (50% modification), 4 indicating 75% opaque (25% modification), and 5 indicating 100% opaque kernels. The kernels with a score of 2–3 were selected and a random sample of 20 seeds per line was sent for laboratory analysis. As a result, all the 58 inbred lines were found to have high tryptophan content (>0.07%) showing that they are all QPM (Appendix 1). Hence, the remaining selected seeds were used for crossing and per se performance evaluation.

4.3.2. SNP analysis

Seedlings of all the inbred lines were raised in pots for about 2 weeks, until they reached 3–4 leaf stage, in a contained room at Addis Ababa University, Addis Ababa. Leaf discs were collected from two leaves of each of the six plants per genotype using leaf cutting tool supplied in KBioscience's plant sample collection kit (KBioscience, UK; http://www.KBioscience.co.uk). The discs were bulked to be added to the supplied 96-well tube storage rack (containing 12 × 8-strip tubes) and sealed by 12 × perforated 8-strip caps. On top of the sealed tubes, desiccant sachet was placed and the rack lid was fixed in place using elastic band; all of which were supplied in the kit. Finally, the prepared tube storage

rack was sealed inside the provided plastic bag. The packaged leaf discs were shipped to LGC Genomics, UK (http://www.lgcgroup.com) for SNP analysis.

DNA was extracted and samples were genotyped by Kbiosciences (Hoddesdon Herts, UK; http://www.KBioscience.co.uk) with 201 SNPs (Appendix 8) recommended by CIMMYT for maize genotyping (Lu *et al.*, 2009; Kassa Semagn *et al.*, 2012; Dao *et al.*, 2014) plus an additional SNP developed and validated by Martha Hernández Rodríguez (2013) for identification of QPM genotypes using Kompetitive Allele Specific PCR (KASP^{TM}) genotyping platform. The SNP-specific KASP Assay mix and the universal KASP Master Mix were added to the samples. The Assay mix contains three assay-specific non-labelled oligos: two allele specific forward primers and one common reverse primer whereas the Master Mix contains the other components required for PCR: the universal fluorescence resonant energy transfer (FRET) cassettes, ROX^{TM} passive reference dye, taq polymerase, free nucleotides and $MgCl_2$ in an optimized buffer solution. The allele-specific primers each harbour a unique tail sequence that corresponds with the universal FRET cassette; one labelled with FAM^{TM} dye and the other with HEX^{TM} dye. To enable the detection of contamination or non-specific amplification, two wells were used as no template controls (NTCs). Following completion of the initial 35 cycles of PCR, the plate was read on a BMG PHERAStar plate reader. This initial read data was visually inspected to assess the progression of the PCR reaction. The plates were then recycled (3 cycles per recycle step) and read after each recycle step. The read data were visually inspected after each recycle step and once they became satisfactory, they reached endpoint of the PCR reaction and were identified as completed. At this stage, Kraken software from LGC Genomics automatically called genotypes for the samples. The plate read data in Kraken were checked twice and the results became ready for analysis.

4.3.3. DNA extraction for ISSR analysis

Seeds of all the 58 inbred lines were raised in pots in MARC greenhouse at room temperature. Young leaves were collected from 3-5 plants at 5–6 leaves stage of seedlings and dried using silica gel. A double extraction protocol modified from triple extraction procedure of Borsch *et al.* (2003) was used for DNA isolation in Addis Ababa University Genetics Laboratory. To state the procedure step by step, the silica-gel-dried leaves were filled into sterile Eppendrof cap with two 5 mm beads and the caps were loaded in a Mixer Mill for 3 minutes at 30 Hz until the samples are thoroughly powdered. If the samples were not ground good enough, they were reloaded for more time in the mill. Then, 700 µl warm (at 65°C) CTAB (2% cetyltrimethylammoniumbromide, 1% polyvinylpyrrolidone (PVP), 100 mM Tris (pH 8), 20 mM EDTA, 1.4 M NaCl) solution that had already been mixed with 28 mM β-Mercapto-ethanol was added. The leaf tissue and extraction buffer was thoroughly mixed by gently inverting the tube and then was incubated for 30 minutes at 65°C, with periodic gentle rocking. After centrifugation of the tubes for 10 minutes at 13000 rpm, the supernatant was discharged and the precipitates were reincubated in the CTAB solution. The CTAB solution was then extracted twice using 600 µl chloroform followed by thorough continuous turning and inverting of the Eppendorf caps for approximately 5 minutes or longer and centrifuging for 10 minutes at 13000 rpm. Thereafter, approximately two third of the solution volume amount of cooled isopropanol (4°C) was added and freezed for more than 2 h at −20°C. Centrifugation was done as above and the liquid was aspirated. The precipitate was washed in 200 µl ethanol (70%) and centrifuged again similarly as above. The ethanol was aspirated and the DNA-pellets were dried at room temperature. Finally, the pellet was re-suspended in 100 µl TE (1x, p.a. grade) and stored at 4°C. The pellets then passed two cleaning steps: washing by 50 µl cooled (4°C) 7.5 M Ammonium acetate (NH_4Ac) solution

and by 3 M Sodium acetate (NaAC) solution. Each cleaning step with salt was followed by precipitating with 100% ethanol, freezing at −20°C for at least 2 h, centrifugation for 35 minutes at 13000 rpm and aspiration of the fluid. Finally, the pellet was re-suspended in 100 µl TE (1x, p.a. grade) and stored at 4°C. The quantity of extracted genomic DNA and its quality were checked after running 7 µl of DNA samples on 1% agarose gel. The DNA concentration was also measured using Thermo Scientific Nano Drop 2000/2000c Spectrophotometer. The resulting DNA concentration was diluted using sterile demineralized water to a concentration of 60–100 ng/ml.

4.3.4. ISSR-PCR analysis

Random samples from each of the putative populations totaling thirty-five inbred lines were used for ISSR analysis. Twenty-four ISSR primers obtained from the University of British Colombia, Primer kit UBC 900, were chosen (Table 2) based on previous studies (Carvalho *et al.*, 2002; do Amaral Júnior *et al.*, 2011; Idris *et al.*, 2012; Nkongolo and Mbuya, 2015) and primer availability. These primers were used for preliminary amplification of twelve randomly selected inbred lines. The amplification reactions were carried out in a final volume of 26 µl containing 120–200 ng of genomic DNA diluted with sterile demineralized water, Taq buffer (10x reaction buffer S) (5 mM Tris-HCl, pH 8.3 [at 25°C]; 25 mM KCl; 0.75 mM $MgCl_2$; 0.89 mM Tween 20), 3 mM $MgCl_2$, 3.8 mM of dNTP mix (0.95 mM of each dTTP, dATP, dCTP and dGTP) (MolBio™ HiMedia®, Mumbai, India), 8 pmol primer and 0.08 units of FIREPol® Taq DNA polymerase (Solis BioDyne, Tartu, Estonia). The final volume was supplemented with 16.7µl double distilled water. The samples were amplified in Biometra® 2003 T3 thermal cycler, including for each primer, a negative control reaction having all the components of the reaction mixture except DNA. The cycles were performed following the program: an initial denaturation for four minutes at 94°C; 40 cycles

(denaturation for 15 seconds at 94°C, primer annealing for one minute at annealing temperatures of respective primers (Table 2), extension for one minute and 30 seconds at 72°C) and a final extension for 7 minutes at 72°C. The resulting PCR products were incubated at 4°C for further activities. The presence or absence of fragments was tested after 3 µl of the product was run on 1% agarose gel. Once presence of band/bands is confirmed, 10 µl of the product was run on more concentrated (1.67%) agarose gel for separation of the fragments.

Table 2. Nucleotide sequence of the ISSR primers used to amplify genomic DNA from thirty-five maize inbred lines

Number	**Primer Name**	**Nucleotide Sequence (5'→ 3')**	**Annealing Temperature (°C)**	**Remark**
1	808	$(AG)_8C$	48	No clear band, rejected
2	809	$(AG)_8G$	48	No clear band, rejected
3	811	$(GA)_8C$	48	No clear band, rejected
4	812	$(GA)_8A$	45	Clear band, selected
5	813	$(CT)_8T$	45	No clear band, rejected
6	814	$(CT)_8A$	45	No clear band, rejected
7	818	$(CA)_8G$	48	No clear band, rejected
8	820	$(GT)_8C$	48	No clear band, rejected
9	822	$(TC)_8A$	45	No clear band, rejected
10	824	$(TC)_8G$	48	Clear band, selected
11	826	$(AC)_8C$	48	No clear band, rejected
12	835	$(AG)_8YC$	48	Clear band, selected
13	836	$(AG)_8YA$**	48	Clear band, selected

Table 2. Nucleotide sequence of the ISSR primers used continued

Number	Primer Name	Nucleotide Sequence (5'→ 3')	Annealing Temperature (°C)	Remark
14	841	$(GA)_8YC$**	53	No clear band, rejected
15	844	$(CT)_8RC$*	48	No clear band, rejected
16	848	$(CA)_8RG$*	48	Clear band, selected
17	854	$(TC)_8RG$*	48	No clear band, rejected
18	857	$(AC)_8YG$**	48	No clear band, rejected
19	872	$(GATA)_4$	38	No clear band, rejected
20	873	$(GACA)_4$	45	Clear band, selected
21	874	$(CCCT)_4$	53	No clear band, rejected
22	878	$(GGAT)_4$	45	No clear band, rejected
23	880	$(GGAGA)_3$	45	Clear band, selected
24	881	$(GGGTG)_3$	53	No clear band, rejected

**R = A or G and **Y = C or T*

4.3.5. Agarose gel electrophoresis

Agarose gel preparation and electrophoresis procedures were modified from Lee *et al.* (2012). Preparation of standard 1% agarose gel is started by adding 0.5 g or 0.835 g electrophoresis grade agarose powder for test gel or fragment separation, respectively to 50 ml of 1X TBE (0.1 mM Tris, 0.1 M boric acid, 2 mM EDTA pH 8.0) inside a conical flask. The agarose-buffer mixture is then melted by heating in a microwave oven and is swirled to ensure even mixing until the agarose was completely dissolved while maintaining adequate thickness. Melted agarose was left on the benchtop for 3–5 minutes to be cooled and 1 ml of 10mg/ml ethidium bromide (EtBr) solution was mixed. The gel is, soon afterward, poured slowly on a gel tray that is already sealed into a casting apparatus to prevent leakage and an

appropriate gel comb was inserted, making sure that no bubbles were trapped underneath the combs and all bubbles on the surface of the agarose were removed before the gel set. Then, it was left to set for at least 30 minutes. After the gel has hardened, the gel comb was withdrawn, taking care not to tear the sample wells. The gel casting platform containing the set gel is placed in an electrophoresis tank (BIO RAD SUB-CELL® GT) that has already been filled with 1X TBE buffer sufficient to submerge the tops of the wells.

In order to run gel electrophoresis, an appropriate amount of DNA samples to be checked or separated were mixed with 2 ml of loading dye and loaded into each well. Gel loading dye was made at 6X concentration (4.1 M Glycerol, 20 mM Tris-Hcl, 3.6 mM Bromophenol Blue). These products were run against 1 kb plus DNA ladder loaded on the first well so as to determine the size of the fragments generated and a negative control loaded on the last well. The cathod (black leads on the lid of the tank) was checked if it is correctly placed closer to the wells than the anode (red leads) and the electrodes were plugged into the correct slots in the power supply (Biometra® Standard Power Pack P25). The supply was programmed at 80 V and turned on until the bromphenol blue dye migrated a distance judged sufficient for DNA quantity and quality analysis or separation of the DNA fragments. The agarose gels were finally visualized and documented by using the BIO RAD Gel Doc ™ EZ Imager.

4.4. Data analysis

4.4.1. Morphological data analysis

4.4.1.1. Analysis of variance

Analysis of variance (ANOVA) was performed for grain yield, yield parameters and other agronomic traits within environments with the PROC MIXED procedure in SAS proprietary software version 9.0 (SAS, 2002) considering genotypes as fixed effects and replications and incomplete blocks within replications as random. F tests were considered significant at $p \leq 0.05$ and highly significant at $p \leq 0.01$. Least significant difference (LSD) was used to compare means at $p \leq 0.05$. Prior to ANOVA, data were tested for homogeneity according to Gomez and Gomez (1984). Based on the test result, all the variances of considered traits were homogeneous.

Individual environment

The effects model used for the ANOVA of each environment was:

$$Y_{ijkl} = \mu + g_i + r_j + b(r)_{jk} + e_{ijk}$$

where μ is the overall mean, g_i is the effect of the i^{th} genotype, is r_j the effect of the j^{th} replication, $b(r)_{jk}$ the effect of the k^{th} incomplete block within the j^{th} replication and e_{ijk} is the residual variance.

Skeleton of ANOVA for individual site is given in Table 3 and Table 4 in which sum of squares due to parents is partitioned into sum of squares due to lines and testers in the lines trials. Orthogonal comparisons of lines versus testers were also made. Likewise, sum of squares due to genotypes were partitioned into hybrid, check and hybrid versus check sum of squares in the hybrids trials.

Table 3. Outline of analysis of variance for parents at each site

Source of variation	**Degree of freedom**	**Mean square**
Replications	$r - 1$	MS_r
Blocks (Replications)	$r(b - 1)$	MS_b
Parents	$p - 1$	MS_p
Lines	$l - 1$	MS_l
Testers	$t - 1$	MS_t
Lines vs. Testers	1	$MS_{l\ vs\ t}$
Error	$(r - 1)(p - 1)$	MS_e
Total	$rp - 1$	MS_T

Table 4. Outline of analysis of variance for hybrids at each site

Source of variation	**Degree of freedom**	**Mean square**
Replications	$r - 1$	MS_r
Blocks (Replications)	$r(b - 1)$	MS_b
Genotypes	$g - 1$	MS_g
Hybrids	$h - 1$	MS_h
Checks	$c - 1$	MS_c
Hybrids vs. checks	1	$MS_{h\ vs\ c}$
Error	$(r - 1)(g - 1)$	MS_e
Total	$rg - 1$	MS_T

Multi-environment

Mean square error variances of each trait for each site were tested for their homogeneity based on Bartlett's test (Gomez and Gomez, 1984) so that data for each trait was combined using PROC GLM in SAS proprietary software version 9.0 (SAS, 2002) across sites. Combined analysis was performed for traits that had homogeneous error variances and

significant differences in the individual site analyses. Genotypes were considered as fixed effects and sites, replications and blocks within replications as random effects. The combined analysis was performed using genotype means adjusted for block effects generated from individual sites analyses according to the lattice design. The linear effects model for the combined analysis of hybrids across sites for all traits of interest was:

$$Y_{ijkl} = \mu + g_i + s_j + (g \times s)_{ij} + r(s)_{jk} + e_{ijkl}$$

where μ is the overall mean, g_i is the effect of the i^{th} genotype, env_j is the effect of the j^{th} site, $(g \times s)_{ij}$ is the interaction effect of the i^{th} genotype by the j^{th} site, $r(s)_{jk}$ is the effect of the k^{th} replication within the j^{th} site and e_{ijkl} is the residual variance.

As indicated in Table 5 and Table 6, parents and their interaction with site sum of squares were partitioned into sum of squares due to lines, testers and lines versus testers contrast and their respective interactions with the sites in the lines trial. Genotype sum of squares were partitioned into variations due to hybrids, checks and the contrast between hybrids and checks and their respective interactions with the sites in the hybrids trial.

Mean squares for parents × environment (E), hybrids × E, lines × E, tester × E and checks × E were considered as error terms to test the significance of mean squares for parents, hybrids, lines, testers and checks, respectively. In the same way, the contrasts between lines and testers, parents and hybrids and hybrids and checks tested against the mean squares for their corresponding interaction with environment as error term.

Whereas, the significance of the mean squares of the interactions with the environment were tested against pooled error mean square as error term that was obtained by dividing the sum of error sum of squares from all locations with the corresponding sum of the error degrees of freedom. Simply, it can be calculated using the equation:

$$Pooled\ error\ mean\ square = \frac{\sum_{j=1}^{e} K_j S_j^2}{\sum_{j=1}^{e} K_j r}$$

In which K_j and S_j^2 represent error degree of freedom and error mean square at j^{th} environment, respectively, while e and r denote the number of environments and the number of replications in each environment, respectively.

Table 5. Outline of analysis of variance for parents across sites

Source of variation	**Degree of freedom**	**Mean square**
Sites (s)	$s-1$	
Replications (Sites)	$s(r-1)$	MS_r
Parents	$p-1$	MS_p
Lines	$l-1$	MS_l
Testers	$t-1$	MS_t
Lines vs testers	1	$MS_{l\ vs\ t}$
Parents × s	$(p-1)(s-1)$	$MS_{p\times s}$
Line × s	$(l-1)(s-1)$	$MS_{l\times s}$
Tester × s	$(t-1)(s-1)$	$MS_{t\times s}$
Line vs tester × s	$1\ (s-1)$	$MS_{(l\ vs\ t)\times s}$
Error	$s(r-1)(p-1)$	MS_e
Total	$srp-1$	MS_T

Table 6. Outline of analysis of variance for hybrids across sites

Source of variation	**Degree of freedom**	**Mean square**
Sites (s)	$s-1$	
Replications (Sites)	$s(r-1)$	MS_r
Genotypes	$g-1$	MS_g
Hybrids	$h-1$	MS_h
Checks	$c-1$	MS_c
Hybrids vs checks	1	$MS_{h\ vs\ c}$
Genotypes × s	$(g-1)(s-1)$	$MS_{g\times s}$
Hybrids × s	$(h-1)(s-1)$	$MS_{h\times s}$
Checks × s	$(c-1)(s-1)$	$MS_{c\times s}$
Hybrids vs checks × s	$1\ (s-1)$	$MS_{(h\ vs\ c)\times s}$
Error	$s(r-1)(g-1)$	MS_e
Total	$srg-1$	MS_T

4.4.1.2. Combining ability analysis

Line × tester (L × T) analysis was performed using the adjusted means for block effects in the individual analyses for traits that showed significant differences among hybrids. The analysis was performed using SAS proprietary software version 9.0 (SAS, 2002) for individual site and combined across sites in order to partition the mean squares due to hybrids into lines, testers and line × tester effects. The L × T analysis was conducted to estimate general combining ability (GCA) effects of the parents and specific combining ability (SCA) effects of the hybrid combinations considering the genotypes as populations (taking genotypes as fixed effects).

i. General Combining Ability (GCA)

General combining ability (GCA) effect of lines and testers is defined as a deviation of line- and tester-mean from mean of hybrids and calculated using the following equations:

a) GCA of lines

$$GCA_i = \frac{X_{i\,.\,.}}{tr} - \frac{X_{.\,.\,.}}{ltr}$$

b) GCA of testers

$$GCA_j = \frac{X_{.\,j\,.}}{lr} - \frac{X_{.\,.\,.}}{ltr}$$

where GCA_i = GCA effect for the i^{th} line with $\sum GCA_i = 0$; GCA_j = GCA effect for the j^{th} tester with $\sum GCA_j = 0$; $X_{i\,.\,.}$ = the total of i^{th} line over all testers (t) and replications (r); $X_{.\,j\,.}$ = the total of the j^{th} tester over all lines (l) and replication (r) and $X_{.\,.\,.}$ = the total of all the hybrids over all lines (l), testers (t) and replications (r).

ii. Specific Combining Ability (SCA)

Specific combining ability (SCA) effect of hybrid combinations is the deviation of each hybrid-mean from the mean of all hybrids adjusted for corresponding GCA effects of parents and is computed as:

$$SCA_{ij} = \frac{X_{i\,j\,.}}{r} - \frac{X_{i\,.\,.}}{tr} - \frac{X_{.\,j\,.}}{lr} + \frac{X_{.\,.\,.}}{ltr}$$

Where SCA_{ij} = SCA effect of the ij^{th} hybrid with $\sum S_{ij} = 0$ for each j; $X_{i\,j\,.}$ = the total of ij^{th} hybrid combination over all replications (r).

iii. Standard errors for combining ability effects

The significance of GCA or SCA effects was tested by dividing the GCA effects of a particular line or tester and SCA effects of a particular hybrid by its respective standard error (SE). Therefore, the SE was computed using SAS proprietary software version 9.0 (SAS, 2002) using the following formulae:

a) Standard errors for GCA

For lines

$$SE(g_i) = \sqrt{\frac{errorMS\ (l-1)}{ltr}}$$

For testers

$$SE(g_j) = \sqrt{\frac{errorMS\ (t-1)}{ltr}}$$

b) Standard error for SCA

$$SE(S_{ij}) = \sqrt{\frac{errorMS\ (l-1)(t-1)}{ltr}}$$

Individual environment

Combining ability analysis for each environment was carried out based on the following linear model:

$$Y_{ijk} = \mu + GCA_i + GCA_j + SCA_{ij} + e_{ijk}$$

where Y_{ijk} is the observed measurement for the ij^{th} hybrid grown in the k^{th} replication or site; μ is the population mean; GCA_i and GCA_j are the GCA effects; SCA_{ij} the SCA effect; and e_{ijk} the error term associated with the ij^{th} hybrid evaluated in the k^{th} replication or site. The skeleton for the combining ability analysis of individual site is given in Table 7.

Table 7. Outline for combining ability analysis for L × T crosses evaluated at individual site

Source of variation	**Degree of freedom**	**Mean square**
Replications	$r-1$	MS_r
Hybrids	$h-1$	MS_h
GCA_{lines}	$l-1$	MS_l
$GCA_{testers}$	$t-1$	MS_t
$SCA_{L\times T}$	$(l-1)(t-1)$	$MS_{l\times t}$
Error	$(r-1)(h-1)$	MS_e
Total	$rh-1$	MS_T

Multi-environment

The GCA effect of lines and testers, the SCA effect of line × tester, and their interactions with the environment were determined assuming the model:

$$Y_{ijk} = \mu + GCA_i + GCA_j + SCA_{ij} + s_k + (GCA \times s)_{ik} + (GCA \times s)_{jk} + (SCA \times s)_{ijk} + \epsilon_{ijk}$$

Where Y_{ijk} = the performance of the hybrid made with ith line and j^{th} tester in the k^{th} site, μ = the overall mean, GCA_i = the GCA effect of the i^{th} line, GCA_j = the GCA effect of the j^{th} tester, SCA_{ij} = the interaction of the i^{th} line with the j^{th} tester (SCA effect of the ij^{th} hybrid), s_k = the effect of the k^{th} site, $(GCA \times s)_{ik}$ = the interaction of the GCA_i and s_k, $(GCA \times s)_{jk}$ = the interaction of the GCA_j and s_k, $(SCA \times s)_{ijk}$ = the interaction of SCA_{ij} and s_k.

Table 8 shows the skeleton for the combining ability analysis across locations. The significance of GCA and SCA sources of variation was determined using GCA × s and SCA × s interactions as error terms, respectively. The significance of GCA × s and SCA × s interactions in turn was determined using the error mean squares obtained by dividing the pooled error mean squares from the ANOVA by the number of replications since the combining ability mean squares were calculated based on entry means. The Student's t-test was used to test the null hypothesis that GCA and SCA effect values are zero.

Table 8. Outline for combining ability analysis for parents and hybrids across environments

Source of variation	**Degree of freedom**	**Mean square**
Sites (s)	$s-1$	
Replications (Environments)	$s(r-1)$	MS_r
Hybrids	$h-1$	MS_h
GCA_{lines}	$l-1$	MS_l
$GCA_{testers}$	$t-1$	MS_t
$SCA_{L\times T}$	$(l-1)(t-1)$	$MS_{l\times t}$
Hybrid × s	$(h-1)(s-1)$	$MS_{h\times s}$
GCA_{Lines} × s	$(l-1)(s-1)$	$MS_{l\times s}$
$GCA_{Testers}$ × s	$(t-1)(s-1)$	$MS_{t\times s}$
$SCA_{L\times T}$ × s	$(l-1)(t-1)(s-1)$	$MS_{l\times t\times s}$
Error	$e(r-1)(h-1)$	MS_e
Total	$srl(t-1)$	MS_T

4.4.1.3. Proportional contribution

Proportional contribution of lines, testers and L × T to the sum of squares of hybrids were computed as follows:

$$Contribution\ of\ lines\ (\%) = \frac{SS_l}{SS_h} \times 100$$

$$Contribution\ of\ testers\ (\%) = \frac{SS_t}{SS_h} \times 100$$

$$Contribution\ of\ line \times tester\ (\%) = \frac{SS_{l\times t}}{SS_h} \times 100$$

where SS_h = Sum of squares due to hybrids; SS_l = Sum of squares due to lines; SS_t = Sum of squares due to testers and $SS_{l\times t}$ = Sum of squares due to line × tester.

4.4.1.4. Components of variability

Phenotypic, genotypic and environmental variances were computed from the respective mean squares as indicated below:

i. Genotypic variance (σ_g^2)

$$\sigma_g^2 = \frac{MS_g - MS_{g\times s}}{rs}$$

where MS_g = mean square of genotype; $MS_{g\times s}$ = mean square due to genotype by site interaction; r = number of replications; s = number of sites

ii. Genotype × site interaction variance ($\sigma_{g\times s}^2$)

$$\sigma_{g\times s}^2 = \frac{MS_{g\times s} - MS_e}{r}$$

where $MS_{g\times s}$ = mean square due to genotype × site interaction; MS_e = error mean square

iii. Phenotypic variance (σ_p^2)

$$\sigma_p^2 = \sigma_g^2 + \frac{\sigma_{g\times s}^2}{s} + \frac{\sigma_e^2}{rs}$$

iv. Genotypic (GCV) and phenotypic (PCV) coefficient of variation

$$GCV = \frac{\sqrt{\sigma_g^2}}{\bar{X}} \times 100$$

$$PCV = \frac{\sqrt{\sigma_p^2}}{\bar{X}} \times 100$$

where $\bar{X}$ = grand mean for the trait considered

4.4.1.5. Heritability/Repeatability

Heritability in broad sense (H_b^2) for each site was calculated as:

$$H_b^2 = \frac{\sigma_g^2}{\sigma_p^2} \times 100 = \frac{\sigma_g^2}{\sigma_g^2 + \frac{\sigma_e^2}{r}} \times 100$$

For across sites

$$H_b^2 = \frac{\sigma_g^2}{\sigma_p^2} \times 100 = \frac{\sigma_g^2}{\sigma_g^2 + \frac{\sigma_{g\times s}^2}{s} + \frac{\sigma_e^2}{rs}} \times 100$$

4.4.1.6. Genetic advance

Genetic advance (GA) was calculated using the formula $I\sigma_p H_b^2$ and presented as percentage of mean (GAM) using the formula $\frac{GA}{\bar{X}} \times 100$. In these equations, $I = 2.06$ when top 5%

individuals are selected; σ_p = Phenotypic standard deviation; H_b^2 = Heritability in broad sense and $\bar{X}$ = overall mean.

4.4.1.7. Heterosis

Mid-parent heterosis (average or relative heterosis) was calculated as:

$$MPH = \frac{(F_1 - MPV)}{MPV} \times 100$$

Better-parent heterosis (heterobeltiosis) was calculated as:

$$BPH = \frac{(F_1 - BPV)}{BPV} \times 100$$

Heterosis over check (standard/economic heterosis) was calculated as:

$$SH = \frac{(F_1 - BCV)}{BCV} \times 100$$

where F_1 is the mean performance of the cross; MPV is $\frac{P_1+P_2}{2}$ where P_1 and P_2 are the means of the two inbred parents; BPV is the mean value of the highest performing (best) parent and BCV is the mean performance of the best check variety in a given character. Since the significance of mid parent, high parent and standard heterosis were tested using t statistic, calculated t values were computed as:

t value for mid-parent heterosis (MPH)

$$t_{MP} = \frac{F_1 - MPV}{\sqrt{\frac{3errorMS}{2r}}}$$

t value for better parent heterosis (BPH)

$$t_{BP} = \frac{F_1 - BPV}{\sqrt{\frac{2errorMS}{r}}}$$

t value for standard heterosis (SH)

$$t_{SH} = \frac{F_1 - BCV}{\sqrt{\frac{2errorMS}{r}}}$$

Finally, the calculated t values were compared to the tabular value of t-test at error degrees of freedom corresponding to 5% or 1% level of significance.

4.4.1.8. Correlation among characters

Genotypic and phenotypic correlation coefficients were worked out to find out the relationship between yield, yield components and agronomic traits using META-R statistical software version 5 and the equations used are:

Genotypic correlation

$$r = \sqrt{\frac{COV_{g1,2}}{\sigma_{g_1}^2 \times \sigma_{g_2}^2}}$$

Phenotypic correlation

$$r = \sqrt{\frac{COV_{p1,2}}{\sigma_{p_1}^2 \times \sigma_{p_2}^2}}$$

Where $COV_{g1,2}$ = Genotypic covariance between two traits (1 and 2); $COV_{p1,2}$ = Phenotypic covariance between two traits (1 and 2); $\sigma_{g_1}^2$ = Genotypic variance for first trait; $\sigma_{g_2}^2$ =

Phenotypic variance for first trait; $\sigma_{g_2}^2$ = Genotypic variance for second trait and $\sigma_{p_2}^2$ = Phenotypic variance for second trait.

4.4.2. Molecular analysis

4.4.2.1. SNP data analysis

Summary statistics, including number of alleles, allele frequency, number of genotypes, genotype frequency, observed heterozygosity, unbiased estimate of gene diversity (D_i), polymorphic information content (PIC) and inbreeding coefficient (f) were computed for each SNP using PowerMarker version 3.25 (Liu and Muse, 2005) based on Weir (1996). PIC refers to the relative value of each marker or locus with respect to the amount of polymorphism exhibited and its expected value was calculated as given by Botstein *et al.* (1980) with modification for the diallelic SNP loci, that is:

$$PIC_i = 1 - (p_i^2 + q_i^2) - 2p_i^2 q_i^2$$

where p_i and q_i are the frequencies of the first (major) and the second (minor) allele for locus i respectively. Gene diversity (also called expected heterozygosity) is defined as the probability that two alleles randomly chosen from the test sample are different. The common biased estimator of the gene diversity for locus i can be obtained using the above equation by dropping the last item as:

$$H = 1 - (p_i^2 + q_i^2)$$

while the unbiased estimator at the i^{th} locus is:

$$D_i = \frac{1 - (p_i^2 + q_i^2)}{1 - \frac{1 + f}{n}}$$

where p_i and q_i are the frequencies of allele 1 and 2 for locus i respectively, n is the number of observations for a marker locus which refers to the number of non-missing genotypes observed in the sample and f is the inbreeding coefficient which is defined as the probability for a randomly chosen locus that the two alleles of the individual are inherited identically by descent from a common ancestor (DeGiorgio *et al.*, 2010) and its estimate for two alleles is given by:

$$f = \frac{(n-1)\frac{n_{pq}}{n}}{2np_iq_i - \frac{n_{pq}}{2n}}$$

where n is the number of observations while npq is the number of heterozygotes with allele 1 and 2 and p_i and q_i are as defined previously. Allele frequency was calculated for each locus across all the lines and statistical significance of differences in allele frequency was based on the *P* value from Fisher (1922) exact test.

PowerMarker was used also to compute matrices of Rogers (1972) genetic distance between each pair of lines in the study. Its formula that was modified for the two alleles of SNP marker loci is given as:

$$D_R = \frac{1}{m}\sum_{i=1}^{m}\sqrt{\frac{1}{2}[(p_{iX} - p_{iY})^2 + (q_{iX} - q_{iY})^2]}$$

in which p_{iX} and q_{iX} are the frequencies of the first and second alleles at the i^{th} locus in population X and p_{iY} and q_{iY} are that of population Y and m is the number of loci examined.

Three types of multivariate analysis methods viz. cluster analysis, model-based population structure analysis and principal component analysis were carried out to subdivide inbred lines into genetic subgroups.

Cluster analysis was performed and a dendrogram was constructed from the Rogers' genetic distance matrix using the Neighbour-Joining (NJ, Saitou and Nei, 1987) algorithm using DARwin software version 6 (Perrier *et al.*, 2003). Groups and subgroups were identified from the resultant phylogenetic tree.

The population structure of the 59 inbred lines was inferred using the model-based STRUCTURE software package version 2.3.4 (Pritchard *et al.*, 2000). The number of subpopulations (k) was varied from 1 to 12, each with three replications, using admixture model and assuming correlated allele frequencies and unlinked loci (Falush *et al.*, 2003) with a burn-in period of 100,000 and 100,000 MCMC (Markov Chain Monte Carlo) replications after burn-in. Membership coefficients (Q values) for each inbred line were estimated to have its memberships in multiple subgroups. Lines with $Q \geq 60\%$ were assigned to the corresponding subpopulation while $Q < 60\%$ in any single group were assigned to a "mixed" group. The most appropriate value of k was estimated using both the log likelihood of the data $LnP(D)$ in the STRUCTURE output which refers to the posterior probability of the data with respect to a given k and the ad hoc statistic Δk (Evanno *et al.*, 2005). Evanno's Δk considers the rate of change of $LnP(D)$ as k increases and also the variance of $LnP(D)$ among repeated runs and tends to be maximum at the true value of k. It is calculated as:

$$\Delta k = M \frac{|L(k-1) - 2L(k) + L(k+1)|}{S[L(k)]}$$

in which $L(k)$ represents the k^{th} $LnP(D)$, M is the mean of three runs, and S is their standard deviation.

Principal component analysis (PCA) was conducted using DARwin software (Perrier *et al.*, 2003). The first two principal components were plotted for visual examination of the clustering pattern of lines.

Finally, an analysis of molecular variance (AMOVA) (Excoffier *et al*., 1992) and F statistics (F_{st}) across all subpopulations and between pairwise subpopulations were performed using Arlequin version 3.5.2.2 (Excoffier and Lischer, 2010) to investigate population differentiations among the subpopulations. Individuals were assigned to subpopulations using the results from population structure analysis, cluster analysis and *a priori* pedigree information. The AMOVA partitioned also the variation within subpopulations and within individuals.

4.4.2.2. ISSR data analysis

After photographing using BIO RAD Gel Doc ^{TM} EZ Imager, the ISSR gels were scored manually as binary data that recorded ”1” for the presence and ”0” for the absence of bands whereas ambiguous bands were recorded as ”?”. Only clearly resolved bands were used in the genetic analysis while faint bands were discarded. The data were then used to analyze the genetic associations among the genotypes. A similarity matrix was constructed using the Numerical Taxonomy and Multivariate Analysis System for personal computers (NTSYS-pc) software version 2.1 (Rohlf, 2000) for all pairwise comparisons according to Jaccard (1908) similarity coefficient:

$$J = \frac{N_{11}}{(N_{11} + N_{10} + N_{01})}$$

where N_{11} is the number of bands shared between population i and j; N_{10} is the number of bands present in population i but absent in j and N_{01} is the total number of bands present in population j but not in i.

The reasons for the preferential use of Jaccard’s coefficient were (i) it gives more weight to matches than mismatches (Sneath and Sokal, 1973), (ii) it is less biased by the occurrence of

a given level of artificial bands (Lamboy, 1994) and (iii) sharing or matching of a band has a direct biological meaning in that it is an estimate of the expected properties of amplification. Using the matrix of genetic similarities, cluster analysis was performed in order to deduce genetic relationships among genotypes and evaluate patterns of genotype clustering by the Unweighted Pair Group Method using Arithmetic Averages (UPGMA, Sneath and Sokal, 1973) algorithm with NTSYS-pc version 2.1 (Rohlf, 2000). Jaccard's coefficient was also used to perform principal coordinates analysis (PCoA) with PAST software version 1.18 (Hammer *et al.*, 2001) that helps visualize patterns of variation among germplasm on two dimensional (2D) plot.

Genetic diversity parameters (Toro and Caballero, 2005) were estimated using POPGENE software version 1.32 (Yeh *et al.*, 1997). These parameters include percentage of polymorphic loci ($P\%$), Nei (1973) gene diversity (h), Shannon (1948) information index (I), observed (N_a) and effective (N_e) number of alleles , coefficient of genetic differentiation between germplasm (G_{st}) and estimate of gene flow (N_m) for each population. Finally, analysis of molecular variation (AMOVA) was carried out to calculate variation among and within population as well as within individuals using Arlequin software version 3.5 (Excoffier and Lischer, 2010).

5. Results

5.1. Analysis of variance and mean performances at individual sites

5.1.1. Hybrid trial

Analysis of variance (ANOVA) for individual sites showed significant differences among the hybrids for all measured traits except for ED at MARC, RL, EPP, PA, ED and RPE at Edo Gojola and PH, RL, EPP and CLR at Mieso (Table 9).

At MARC, the minimum GY was 2.67 t ha^{-1} and the maximum 5.68 t ha^{-1} (L53/CML159) with a mean of 4.11 t ha^{-1}. The minimum GY at Edo Gojola was 2.14 t ha^{-1} and the maximum 6.34 t ha^{-1} (L52/CML159) with a site mean of 4.22 t ha^{-1} mean. The minimum and maximum GY at Mieso was 2.46 t ha^{-1} and 5.56 t ha^{-1} (L20/CML159) with a mean yield of 3.97 t ha^{-1}. DA ranged from 65 to 75 days at MARC with a mean value of 70 days while it ranged from 68 to 80 days with a mean value of 75 at Edo Gojola. At MARC, ASI ranged from -3 to 4 days, with a mean value of 0.5 while at Edo Gojola, it ranged from -2 to 7 days with a mean value of 1.8 days. The minimum PH and EH at MARC were 207.8 and 95.0 cm, respectively and the respective maximum values were 256.8 cm and 142.5 cm. Mean PH was 233.3 cm and EH was 119.1 cm. The minimum and maximum PH at Edo Gojola were 190.7 cm and 255.5 cm, respectively, with a mean of 220.8 cm. At the same site, the minimum and maximum EH were 80.7 cm and 145.0 cm, respectively, with a mean of 114.2 cm. Mean values for PH and EH at Mieso were 175.9 and 78.7 cm, respectively, with PH ranging from 149.0 to 196.9 cm and the EH ranging from 51.9 cm to 101.0 cm. EPO ranged from 0.4 to 0.6 with a mean of 0.5 at all the three sites, except at Mieso where the minimum was 0.3. Mean values for EL were 152.11 mm at MARC; 151.59 mm at Edo Gojola and 145.76 mm at Mieso. It ranged from 130.98 to 181.76 mm at MARC, from 126.71 to 179.79 mm at Edo

Table 9. Means, F-test and coefficient of variation (CV%) for grain yield and other agronomic traits of QPM hybrids

Sites	Statistics	GY	DA	ASI	PH	EH	EPO	RL	SL	EPP	CLR	EA	PA	ED	EL	KPR	RPE	TKW
Melkassa	Grand Mean	4.11	69.64	0.50	233.13	119.04	0.51	2.13	2.67	1.03	2.47	2.80	2.44	43.20	152.13	35.35	14.89	195.46
	Minimum	2.67	65.09	-2.52	207.81	95.00	0.40	-0.19	-0.24	0.71	1.75	2.03	2.00	39.27	130.98	29.67	13.17	148.03
	Maximum	5.68	74.96	3.57	256.79	142.50	0.61	18.25	19.50	1.50	2.75	3.55	2.75	53.97	181.76	41.20	17.34	245.15
	Mean of hybrids	4.11	69.61	0.47	233.28	119.05	0.51	2.09	2.68	1.02	2.47	2.79	2.44	43.16	152.11	35.41	14.89	195.45
	Mean of checks	4.13	70.38	1.13	228.75	118.75	0.52	3.85	2.68	2.68	2.25	2.88	2.38	44.29	152.69	33.79	14.94	195.77
	F test	*	**	**	**	**	**	**	**	**	*	**	**	ns	**	**	**	**
	LSD (0.05)	1.21	2.23	2.19	19.28	18.71	0.07	5.14	9.22	0.25	0.37	0.65	0.39	6.66	16.27	3.82	1.23	35.12
	SE(m)	0.61	1.12	1.10	9.71	9.43	0.04	2.59	4.65	0.12	0.19	0.33	0.20	3.36	8.20	1.92	0.62	17.69
	CV %	14.78	1.61	222.43	4.17	7.92	7.05	121.22	173.86	12.13	7.60	11.63	8.00	7.77	5.39	5.44	4.17	9.05
Edo Gojola	Grand Mean	4.18	75.25	1.80	220.85	114.10	0.52	0.17	17.41	0.93	2.43	2.34	2.38	46.07	151.62	34.16	14.86	195.39
	Minimum	2.14	68.36	-1.77	190.72	80.70	0.39	-0.21	-3.78	0.70	1.49	1.39	2.00	40.70	126.71	29.14	13.55	132.95
	Maximum	6.34	80.36	7.02	255.54	144.98	0.63	4.22	78.71	1.35	3.04	3.55	2.75	65.35	179.79	38.79	16.22	261.91
	Mean of hybrids	4.22	75.22	1.82	220.80	114.18	0.52	0.17	17.30	0.93	2.44	2.34	2.39	45.91	151.59	34.19	14.86	194.74
	Mean of checks	4.09	75.88	1.38	222.50	111.88	0.50	0.36	21.62	1.03	2.13	2.38	2.25	50.66	152.47	33.38	14.95	214.12
	F test	*	**	**	*	**	**	ns	**	ns	**	*	ns	ns	**	**	ns	**
	LSD (0.05)	1.52	2.48	2.27	24.51	24.60	0.09	1.96	26.94	0.22	0.48	0.91	0.45	8.65	18.50	4.10	1.36	47.40
	SE(m)	0.77	1.25	1.15	12.35	12.40	0.05	0.99	13.57	0.11	0.24	0.46	0.23	4.36	9.32	2.07	0.69	23.88
	CV %	18.33	1.66	63.52	5.59	10.86	9.15	569.68	77.98	11.73	10.00	19.72	9.54	9.47	6.15	6.05	4.61	12.22
Mieso	Grand Mean	3.97	-	-	176.13	78.77	0.45	1.44	4.24	1.01	2.01	2.82	2.32	42.11	145.99	32.52	14.00	225.57
	Minimum	2.46	-	-	148.96	51.94	0.30	0.00	-2.94	0.74	1.39	2.17	1.73	35.87	121.91	25.97	12.20	164.11
	Maximum	5.56	-	-	196.88	101.01	0.59	15.65	56.38	1.50	2.35	3.51	2.86	52.28	167.59	37.22	15.60	323.20
	Mean of hybrids	3.94	-	-	175.91	78.71	0.45	1.48	4.38	1.01	2.01	2.82	2.31	42.13	145.76	32.48	14.00	224.77
	Mean of checks	4.71	-	-	182.50	80.63	0.44	0.40	0.00	1.12	2.00	2.69	2.44	41.54	153.87	33.50	14.15	247.22
	F test	**	-	-	ns	*	**	ns	**	ns	ns	**	**	**	**	*	**	**
	LSD (0.05)	1.09	-	-	21.24	20.29	0.10	6.66	17.29	0.29	0.57	0.60	0.48	3.49	16.94	4.57	1.33	45.43
	SE(m)	0.55	-	-	10.70	10.22	0.05	3.35	8.71	0.15	0.29	0.30	0.24	1.76	8.54	2.30	0.67	22.88
	CV %	13.86	-	-	6.08	12.98	11.59	233.02	205.68	14.37	14.27	10.66	10.36	4.17	5.85	7.08	4.79	10.15

* *P*< 0.05; ** *P*< 0.01; ns= nonsignificant; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); ED= ear diameter; EH= ear height (cm); EL=ear length; EPO= ear position; EPP= ears per plant; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RL= root lodging; RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean; LSD=least significant difference at 0.05 significance level; CV=coefficient of variation (%)

Gojola and from 121.91 to 167.59 mm at Mieso. The minimum and maximum KPR were 29.67 and 41.20 at MARC, 29.14 and 38.79 at Edo Gojola and 25.97 and 37.22 at Mieso. Similarly, the minimum and maximum RPE were 13.17 and 17.34 at MARC, 13.55 and 16.22 at Edo Gojola and 12.20 and 15.60 at Mieso. Mean KPR was 35.41 at MARC, 34.19 at Edo Gojola and 32.48 at Mieso while respective mean RPE was 14.89, 14.86 and 14.00. TKW ranged from 148.03 to 245.15 g at MARC with a mean of 195.45 g while ranging from 132.95 to 261.91 g at Edo Gojola with a mean of 194.74 g. At Mieso, it ranged from 164.11 to 323.20 g with a mean value of 224.77 g.

5.1.2. Inbred line trial

For the inbred line trial, analysis of variance revealed significant variations among the inbreds for most traits at the three testing sites (Table 10). However, non-significant differences were observed for EH, PA, ED and CLR at MARC, for EA at Edo Gojola and EPP and CLR at Mieso. Mean square for RL was non-significant at all the three sites.

Among the inbred lines evaluated, GY ranged from 0.69 to 3.57 t ha^{-1} at MARC, from 0.77 to 4.31 t ha^{-1} at Edo Gojola and from 0.08 to 3.56 t ha^{-1} at Mieso. Mean GY performances were 1.38, 1.88 and 1.11 t ha^{-1} at MARC, Edo Gojola and Mieso, respectively. The best performing five inbred lines were L52, L53, L38, L47 and L40 at MARC; L52, L47, L48, L17 and L38 at Edo Gojola and L38, L47, L17, L13 and L33 at Mieso. Minimum DA at MARC was 61.59 days while the maximum was 82.44 days with a mean of 70.9 days. At Edo Gojola, DA ranged from 65.93 to 85.3 days with a mean of 73.99 days. At MARC, ASI ranged from -3.05 to 3.7 days with a mean of 0.58 days while it ranged from -4.17 to 5.11 days at Edo Gojola with mean of 1.41 days at Edo Gojola. Mean PH was 158 cm with a range

of 127.88 to 190.18 cm at MARC. The same trait ranged from 119.1 to 199.1 cm with a mean of 157 cm at Edo Gojola, whereas it ranged from 84.92 to 143.46 cm with a mean of 114 cm at Mieso. At Edo Gojola and Mieso, EH ranged from 50.03 to 102.04 cm and 28.86 to 72.09 cm with respective mean values of 76.21 and 44.67 cm. The respective mean, minimum and maximum EPO values were 0.5, 0.36 and 0.63 at MARC; 0.49, 0.35 and 0.66 at Edo Gojola and 0.39, 0.28 and 0.53 at Mieso. SL ranged from -1.52% to 24.36% at MARC with a mean value of 4.48%, from -4.7% to 62.46% at Edo Gojola with a mean value of 16.01% and from -0.91% to 68.12% at Mieso with a mean value of 8.01%. The minimum and maximum EPP values were 0.35 and 1.58 at MARC and 0.53 and 1.51 at Edo Gojola with mean values of 0.83 at MARC and 0.84 at Edo Gojola. At MARC, mean EA was 2.58 ranging from 1.6 to 3.72, whereas at Mieso, mean EA was 3.72 ranging from 2.09 to 4.97. PA had mean values of 2.69 and 3.32 at Edo Gojola and Mieso with ranges of 1.85 to 3.69 and 2.45 to 4.35, respectively. EL ranged from 74.22 to 140.83 mm at MARC, 79.7 to 145.55 mm at Edo Gojola and 66.56 to 132.84 at Mieso with means of 110.91 mm, 110.5 mm and 101.54 mm, respectively. Mean values for numbers of KPR and RPE were 22.91 and 12.84 mm, respectively at MARC, 22.88 mm and 13.27 mm, respectively at Edo Gojola and 20.34 and 12.54 mm, respectively at Mieso. KPR ranged from 15.32 to 28.58 mm at MARC, 13.9 to 32.52 mm at Edo Gojola and 13.52 to 28.1 mm at Mieso. RPE ranged from 9.79 to 15.04 mm at MARC, from 9.68 to 15.15 mm at Edo Gojola and from 9.97 to 15.31 mm at Mieso. TKW ranged from 106.5 and 229.05 g with a mean of 165.39 g at MARC, and from 121.37 to 246.23 g with a mean of 184.61 g at Edo Gojola while it ranged from 119.77 to 238.09 g with a mean of 167.21 g at Mieso.

Table 10. Means, F-test and coefficient of variation (CV%) for grain yield and other agronomic traits of QPM inbred

Sites	Statistics	GY	DA	ASI	PH	EH	EPO	RL	SL	EPP	CLR	EA	PA	ED	EL	KPR	RPE	TKW
Melkassa	Mean	1.38	70.90	0.58	158.00	78.04	0.50	5.51	4.48	0.83	2.59	2.58	2.75	36.19	110.91	22.91	12.84	165.39
	Minimum	0.69	61.59	-3.05	127.88	57.84	0.36	0.00	-1.52	0.35	2.02	1.60	2.18	31.12	74.22	15.32	9.79	106.50
	Maximum	3.57	82.44	3.70	190.18	106.81	0.63	18.00	24.36	1.58	3.23	3.72	3.45	53.62	140.83	28.58	15.04	229.05
	Mean of Lines	1.39	70.53	0.60	157.81	77.78	0.50	5.36	4.62	0.83	2.59	2.57	2.75	36.23	110.48	22.83	12.83	165.95
	Mean of Testers	1.13	81.62	-0.05	163.58	85.56	0.52	9.80	0.30	0.92	2.38	2.90	2.93	34.94	122.95	25.31	13.05	149.20
	F test	**	**	**	**	ns	*	ns	**	**	ns	**	ns	ns	**	**	**	**
	LSD (0.05)	0.78	1.78	2.09	15.47	20.06	0.11	11.36	10.44	0.35	0.63	0.67	0.62	7.71	13.12	3.68	1.32	37.36
	SE(m)	0.38	0.88	1.03	7.64	9.90	0.06	5.60	5.15	0.17	0.31	0.33	0.31	3.80	6.46	1.81	0.65	18.40
	CV %	27.75	1.24	179.50	4.83	12.68	11.22	101.64	115.05	20.95	12.01	12.89	11.08	10.50	5.83	7.90	5.05	11.13
Edo Gojola	Mean	1.88	73.99	1.41	157.04	76.21	0.49	2.88	16.01	0.84	2.67	2.58	2.69	38.16	110.50	22.88	13.27	184.61
	Minimum	0.77	65.93	-4.17	119.10	50.03	0.35	-2.88	-4.70	0.53	1.96	1.25	1.85	33.15	79.70	13.90	9.68	121.37
	Maximum	4.31	85.30	5.11	199.10	102.04	0.66	30.69	62.46	1.51	3.26	4.00	3.69	56.71	145.55	32.52	15.15	246.23
	Mean of Lines	1.86	73.66	1.46	156.82	75.98	0.49	2.95	16.22	0.83	2.69	2.55	2.70	38.13	109.75	22.77	13.25	185.35
	Mean of Testers	2.39	83.51	0.05	163.33	82.83	0.50	0.80	9.97	1.17	2.05	3.38	2.52	38.84	132.30	26.16	13.60	163.01
	F test	**	**	**	**	**	**	ns	**	**	*	ns	**	*	**	**	**	**
	LSD (0.05)	0.74	2.09	2.48	24.31	12.47	0.08	15.39	19.57	0.26	0.61	1.48	0.58	6.64	17.54	3.01	1.24	42.35
	SE(m)	0.37	1.03	1.22	12.00	6.15	0.04	7.57	9.63	0.13	0.30	0.73	0.28	3.27	8.65	1.48	0.61	20.90
	CV %	19.50	1.40	86.74	7.64	8.08	8.11	262.86	60.14	15.49	11.21	28.27	10.55	8.58	7.83	6.49	4.60	11.32
Mieso	Mean	1.11	-	-	114.10	44.67	0.39	5.89	8.01	0.83	2.53	3.72	3.32	34.71	101.54	20.34	12.54	167.21
	Minimum	0.08	-	-	84.92	28.86	0.28	-0.51	-0.91	0.43	2.23	2.09	2.45	29.58	66.56	13.52	9.97	119.77
	Maximum	3.56	-	-	143.46	72.09	0.53	35.60	68.12	1.19	3.47	4.97	4.35	40.71	132.84	28.10	15.31	238.09
	Mean of Lines	1.11	-	-	113.93	44.65	0.39	5.99	8.25	0.83	2.53	3.70	3.32	34.72	101.20	20.28	12.55	167.51
	Mean of Testers	1.11	-	-	118.95	45.15	0.38	2.95	1.23	0.98	2.52	4.28	3.16	34.30	111.60	21.95	12.39	158.52
	F test	**	-	-	**	**	**	ns	**	ns	ns	**	**	**	**	**	*	**
	LSD (0.05)	0.80	-	-	19.29	13.89	0.09	14.50	14.22	0.36	0.39	0.85	0.60	4.65	17.46	4.30	1.76	33.66
	SE(m)	0.39	-	-	9.49	6.84	0.05	7.12	7.00	0.18	0.19	0.42	0.30	2.29	8.61	2.12	0.87	16.60
	CV %	35.49	-	-	8.32	15.31	11.59	120.97	87.32	21.32	7.63	11.29	8.93	6.61	8.48	10.42	6.90	9.93

* *P*< 0.05; ** *P*< 0.01; ns= nonsignificant; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); ED= ear diameter; EH= ear height (cm); EL=ear length; EPO= ear position; EPP= ears per plant; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RL= root lodging; RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean; LSD=least significant difference at 0.05 significance level; CV=coefficient of variation (%)

5.2. Analysis of variance and mean performances across sites

5.2.1. Hybrid trials

Combined analysis of variance for the hybrids showed highly significant differences (<0.01) among sites and genotypes (hybrids and checks) for all measured traits except CLR for site (Table 11). However, no significant differences were observed for genotype × site interaction effects for most considered traits except for GY, SL, EL, KPR, CLR and PA. Partitioning of genotype sum of squares into hybrids, checks and hybrids versus checks contrast revealed highly significant (<0.01) differences among hybrids for all traits but non-significant differences among checks for most traits except DA, EH, EPO, CLR, PA and TKW and non-significant differences for the contrast between hybrids and checks except CLR and TKW. The hybrid × site interaction effects, as the genotype × site interaction effects, were significant only for GY, SL, EL, KPR, CLR and PA. However, non-significant check × site interaction and its contrast with hybrids were observed for all traits except KPR for check × site interaction. Across sites, the highest mean grain yield was observed for L52/CML159 (5.38 t ha^{-1}) followed by L18/CML159 (5.07 t ha^{-1}) and L35/CML159 (5.02 t ha^{-1}). The best standard check was MH140 yielding 4.5 t ha^{-1} followed by MH130 yielding 4.43 t ha^{-1} and L52/CML159 was the only one with significantly higher yield than MH140. However, there were 22 hybrids which yielded statistically equivalent to MH140 and there were 32 hybrids that were higher yielder than the best QPM check, MHQ138 (4.37 t ha^{-1}). Mean DA ranged from 66.7 days for MH130, which is the earliest cultivar released for drought stressed areas, to 77.7 days for BHQPY545 which is a QPM cultivar released for mid-altitude high potential areas. The hybrids L38/CML159 (68.6 days) and L22/CML159 (68.8 days) had male flowering comparable to MH130. The highest yielding standard check, MH140 had DA of 73.8 days while almost all hybrids with comparable or higher yield were significantly earlier. Mean PH ranged from 206.7 cm for L56/CML144 to 245.6 cm

Table 11. Combined analysis of variance and means for grain yield and other agronomic traits of QPM hybrids

SoV	df	GY	EH	EPO	SL	EA	EL	KPR	TKW	df	DA	ASI	PH	CLR	PA	RPE
Site (S)	2	4.00**	115782.5**	0.36**	15868.8**	17.77**	2767.06**	486.34**	72698.37**	1	3774.4**	205.41**	18068.80**	0.17	1.88**	95.14**
Rep(site)	3	15.92**	225.21	0.01*	949.17**	0.51	203.85	34.88**	11542.91**	2	8.85*	32.12**	59.64	0.26**	0.40**	0.15
Genotypes (G)	119	1.57**	507.26**	0.01**	529.84**	0.54**	283.00**	14.60**	2660.82**	119	9.58**	5.41**	372.64**	0.12**	0.10**	1.24**
Hybrids (H)	115	1.59**	486.46**	0.01**	546.33**	0.54**	287.55**	15.01**	2562.16**	115	7.55**	5.58**	376.36**	0.10**	0.10**	1.28**
$GCA_{Line\ (L)}$	57	1.43**	700.82**	0.01**	904.13**	0.72**	472.51**	19.54**	1821.13**	57	9.04**	8.47**	490.41**	0.13**	0.14**	1.84**
$GCA_{Tester\ (T)}$	1	49.13**	6156.47**	0.17**	1448.59**	0.6	80.68	9.81	138532.8**	1	220.69**	22.42**	119.02	0.08	0.19	1.57
$SCA_{L \times T}$	57	0.92	172.62	0.00*	172.71*	0.37**	106.22	10.56*	917.74	57	2.31	2.39*	266.83	0.07*	0.07	0.71*
Check (C)	3	0.90	1473.61**	0.01**	74.08	0.65	123.67	2.35	5806.91**	3	88.08**	0.50	343.75	0.60*	0.14*	0.21
H vs C	1	1.04	1.23	0.00	0.02	0.00	237.86	5.18	4568.04*	1	7.73	0.17	30.82	1.12**	0.01	0.15
G × S	238	0.89*	161.28	0.00	167.55**	0.26	150.34*	9.59**	789.96	119	2.27	1.85	217.86	0.07*	0.08*	0.52
H × S	230	0.87*	163.18	0.00	170.36**	0.26	153.65*	9.31*	796.48	115	2.21	1.88	219.95	0.07*	0.08*	0.54
$GCA_L \times S$	114	0.91	184.33*	0.00	234.46**	0.25	147.24	8.73	768.44	57	1.80	1.74	232.29	0.08*	0.09**	0.42
$GCA_T \times S$	2	0.38	246.12	0.01**	186.88	0.44	345.97	9.92	1688.47	1	9.97*	0.70	19.45	0.01	0.29*	4.29**
$SCA_{L \times T} \times S$	114	0.85	140.57	0.00	105.97	0.27	156.68*	9.88*	808.86	57	2.49	2.04	211.12	0.06	0.06	0.59
C × S	6	1.33	130.90	0.00	67.15	0.32	26.68	18.92*	434.11	3	5.17	0.08	160.42	0.02	0.02	0.03
H vs C × S	2	1.87	34.53	0.00	145.57	0.10	141.35	14.10	1107.89	1	0.05	4.63	150.00	0.03	0.15	0.04
Error for hybrids	345	0.71	143.17	0.00	113.86	0.22	118.32	7.08	673.12	230	2.17	1.52	215.75	0.05	0.05	0.47
Error for checks	9	1.03	125.69	0.00	42.39	0.19	129.25	4.30	405.47	6	1.46	0.29	133.33	0.06	0.02	0.68
Pooled Error	357	0.72	142.02	0.00	111.81	0.23	117.91	7.06	664.23	238	2.14	1.48	212.47	0.05	0.05	0.47
Mean of hybrids		4.09	103.98	0.49	8.12	2.65	149.82	34.03	204.96		72.42	1.15	233.28	2.46	2.38	14.45
Mean of Checks		4.31	103.75	0.49	8.10	2.65	153.01	33.55	219.04		73.13	1.25	211.25	2.13	2.35	14.68
Grand Mean		4.09	103.97	0.49	8.10	2.65	149.91	34.01	205.47		72.44	1.15	226.99	2.45	2.38	14.45
Minimum		3.06	80.86	0.39	-0.27	2.03	136.99	30.71	157.70		66.72	-1.14	206.69	1.62	1.87	13.19
Maximum		5.38	124.45	0.58	42.62	3.35	168.07	37.74	259.05		77.66	4.77	245.58	2.88	2.68	16.47
LSD (0.05)		0.74	12.33	0.05	11.10	0.42	9.97	2.41	24.82		1.67	1.58	15.59	0.30	0.31	0.91
SE(m)		0.65	10.76	0.05	9.69	0.37	8.70	2.10	21.66		1.19	1.12	11.11	0.22	0.22	0.65
% GCA_L		44.43	71.41	66.34	82.03	65.59	81.45	64.54	35.23		59.38	75.25	64.59	62.68	66.03	71.53
% GCA_T		26.84	11.01	16.26	2.31	0.97	0.24	0.57	47.02		25.43	3.50	0.27	0.68	1.66	1.07
% $SCA_{L \times T}$		28.73	17.59	17.41	15.67	33.44	18.31	34.89	17.75		15.19	21.25	35.14	36.64	32.31	27.4

* $P< 0.05$; ** $P< 0.01$; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean; LSD=least significant difference at 0.05 significance level; CV=coefficient of variation (%)

for L37/CML159 while EH ranged from 80.9 cm for L38/CML159 to 124.4 cm for L21/CML144. The hybrid L38/CML159 had the smallest mean EPO. Most of the top 20 higher yielding hybrids were taller with lower ear height than MH140 and smaller EPO. The best tolerant hybrid against SL was L55/CML159 (-0.3%) while the most susceptible one was L23/CML144 (42.6%). All of the top 20 high yielding hybrids were SL tolerant except L21/CML159. The hybrid with the lowest CLR score was MH140 (1.6). The best EA was observed for L47/CML144 (2.0) while 25 hybrids had ear aspect score of less than 2.5. The best PA was scored for L13/CML159 (1.9) and most of the hybrids had PA score of less than 2.5. The hybrid L6/CML159 (52.28 mm) had the thickest ear while L19/CML159 (168.07 mm) had the longest ear followed by L34/CML159 (166.02 mm) and L19/CML144 (164.97 mm). The maximum KPR was recorded for L45/CML144 (37.74) while the minimum was recorded for L53/CML159 (30.71). Mean RPE ranged from 13.19 (L34/CML144) to 16.47 (L47/CML144) and mean TKW ranged from 157.7 g (L43 x CML144) to 259.05 g (L36/CML159). The best TKW was measured also for hybrids L57 x CML159 (246.1 g) and MH130 (245.22 g).

5.2.2. Inbred line trials

Analysis of variance across sites for inbred line trials revealed highly significant differences among sites and parents for all studied traits except EPP for sites (Table 12). Parent × site interaction effects were significant for all observed traits except for EPO, EL, KPR, RPE, ASI and ED. Partitioning of sum of squares for parents into their components showed highly significant differences among lines for all considered traits while non-significant differences were detected among testers and lines versus testers contrast for all studied traits except GY, PH and TKW for testers and EL, KPR, DA and EPP for the contrast. Significant differences among line × site interaction effects were observed for GY, PH, SL, DA, EH, EPP, EA and

PA. However, non-significant tester × site and line versus tester × site interaction effects were observed for all measured traits.

According to across sites analysis, L52 showed the highest mean GY which was 3.15 t ha^{-1} followed by L38 (2.94 t ha^{-1}), L47 (2.88 t ha^{-1}), L17 (2.43 t ha^{-1}), L48 (2.33 t ha^{-1}) and L40 (2.06 t ha^{-1}). These lines were superior to the best inbred line check, CML144 (2.02 t ha^{-1}). All of these lines were also among the earliest lines.

The smallest mean DA was observed for L38 (63.8 days) while L52, L47, L17, L48 and L40 showing DA values of 67.9, 68.8, 64.4, 69.3 and 70.1 days, respectively. They were significantly earlier than both check inbred lines, CML144 (83.9 days) and CML159 (81.3 days).

The line with the lowest PH was L10 (112.0 cm) and with the highest PH was L35 (170.9 cm). The lowest EH was observed for L38 (42.8 cm) while the highest for L33 (77.5 cm). Among the top yielding lines, L38 and L47 had PH (162.9 cm and 163.0 cm, respectively) comparable to the check with higher PH, CML144 (158.9 cm) and they had EH (52.6 cm and 60.2 cm, respectively) comparable to the check with lower EH, CML159 (53.5 cm). L38 had the smallest EPO (0.34) among all the lines while L40 (0.4) had smaller EPO as compared to CML144 (0.5) and CML159 (0.44).

The mean SL ranged from -0.6% (L22) to 41.5% (L27). All of the top yielding nine lines were SL tolerant having SL of less than 8%. The best EPP was recorded for CML144 (1.36) followed by L40 (1.24) and L52 (1.07) which were among the top six high yielding inbred lines. All of the top six high yielding lines had the best EA scores than both checks.

They also had the best yield components viz. ED, EL, KPR, RPE and TKW and were superior to both checks in terms of these traits.

Table 12. Combined analysis of variance and means for grain yield and other agronomic traits of QPM inbred lines

SoV	df	GY	PH	EPO	SL	EL	KPR	RPE	TKW	df	DA	ASI	EH	EPP	EA	PA	ED
Site (S)	2	17.49**	74139.62**	0.4**	3950.16**	3358.05**	262.33**	14.46**	12913.44**	1	555.27**	39.05**	58795.02**	0.01	76.0**	21.9**	690.1**
Rep(Site)	3	6.53**	4190.54**	0.0**	69.59	1586.14**	136.05**	9.46**	9226.68**	2	15.22**	4.79*	1045.87**	0.5**	1.35**	4.10**	51.72**
Parent (P)	58	1.59**	1160.70**	0.0**	781.06**	840.70**	55.33**	3.79**	3240.22**	58	39.58**	11.43**	348.95**	0.0**	0.99**	0.51**	31.64**
Line (L)	56	1.60**	1173.95**	0.0**	801.39**	821.16**	54.08**	3.91**	3161.55**	56	25.68**	11.55**	341.08**	0.0**	0.99**	0.52**	32.75**
Tester (T)	1	2.64*	1008.33*	0.01	0.16	214.12	75.35	0.65	8275.58*	1	12.50	8.00	1128.13	0.81	1.13	0.00	0.05
L vs T	1	0.12	570.92	0.00	423.35	2561.51**	105.17**	0.67	2610.34	1	845.00**	7.87	10.64	0.2*	1.02	0.50	0.72
P x S	116	0.40**	213.61**	0.00	201.70**	94.41	6.14	0.55	576.45*	58	2.63**	1.79	124.98*	0.0*	0.54**	0.27*	10.07
L x S	112	0.40**	218.18**	0.00	208.60**	91.83	6.16	0.54	545.9	56	2.60**	1.83	122.40*	0.0*	0.56**	0.28*	10.33
T x S	2	0.02	164.58	0.00	10.85	168.64	7.44	1.49	2539.88	1	4.50	0.00	378.13	0.00	0.00	0.00	1.89
L vs T x S	2	0.86	6.98	0.00	5.65	165.14	3.80	0.26	323.77	1	2.36	1.37	16.44	0.17	0.03	0.03	3.32
Error	174	0.25	128.61	0	71.83	91.75	5.6	0.66	411.51	116	1.23	1.49	80.35	0.03	0.23	0.17	9.20
Mean of lines		1.44	142.85	0.46	9.57	107.04	21.94	12.89	172.94		72.10	1.03	60.31	0.83	3.14	3.01	36.43
Mean of testers		1.55	148.62	0.47	3.83	122.28	24.47	13.01	156.91		82.57	0.00	63.99	1.04	3.59	2.84	36.57
Grand Mean		1.45	143.0	0.46	9.4	107.55	22.02	12.89	172.40		72.4	1.0	60.4	0.83	3.2	3.0	36.43
Minimum		0.54	112.0	0.34	-0.6	79.68	14.57	9.81	128.23		63.8	-2.8	42.8	0.57	1.8	2.4	31.72
Maximum		3.15	170.9	0.58	41.5	138.90	29.65	15.14	225.13		83.9	3.8	77.5	1.36	4.1	3.8	47.38
LSD (0.05)		0.45	11.5	0.06	8.8	9.33	2.13	0.84	21.89		1.4	1.6	9.3	0.22	0.5	0.4	4.05
SE(m)		0.38	9.87	0.05	7.49	7.97	1.82	0.72	18.72		0.96	1.13	6.50	0.15	0.38	0.29	2.83

* *P*< 0.05; ** *P*< 0.01; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); ED= ear diameter; EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean; LSD=least significant difference at 0.05 significance level

5.3. Combining ability analysis

Combining ability analysis indicated significant line GCA mean squares for all measured traits. Mean squares attributable to tester GCA were significant only for GY, EH, EPO, SL, TKW, DA and ASI while line × tester interactions were significant only for EPO, SL, EA, KPR, ASI, CLR and RPE. The line GCA × site interaction effects were significant only for EH, SL, CLR and PA and tester GCA × site interaction effects were significant for EPO, DA, PA and RPE while SCA × site interaction effects were significant for EL and KPR (Table 11).

5.3.1. General combining ability effects

General combining ability (GCA) effects across sites are presented in Table 13. GCA effects of the 58 lines and two testers were found to be very variable for all the 17 traits studied. GCA for GY varied from -0.61 t ha^{-1}(L11) to 0.83 t ha^{-1} (L35). Inbred lines L35 (0.83 t ha^{-1}) and L45 (0.68 t ha^{-1}) had highly significant ($P < 0.01$) positive GCA effects and L53 (0.63 t ha^{-1}), L4 (0.57 t ha^{-1}), L21 (0.56 t ha^{-1}), L52 (0.54 t ha^{-1}) and L32 (0.49 t ha^{-1}) had significant ($P < 0.05$) and positive GCA effects for GY.

Inbred lines L22 (-1.79 days), L39 (-1.79 days), L38 (-1.54 days) L45 (-1.42 days), L27 (-1.29 days), L51 (-1.29 days), L58 (-1.29 days), L12 (-1.17 days) and L52 (-1.17 days) were the best general combiners for earliness with highly significant negative GCA effects for DA. GCA value for ASI for L32, L35, L36 and L54 was -1.65 days, which was highly significant. In addition, several other lines L40 and L50 (-1.52 days), L49 (-1.40 days), L56 (-1.27 days), L51 (-1.15 days) and L58 (-1.15 days) showed negative and highly significant GCA effects for ASI.

GCA for shorter plant stature was exhibited by L56 (-18.91 cm), L55 (-15.16 cm) and L8 (-12.66 cm) whereas inbred lines L21 (16.09 cm), L20 (14.21 cm), L19 (13.59 cm) and L35 (12.96 cm) showed a tendency to increase plant height. Inbred lines with the lowest GCA effects

for ear height was L45 (-13.15 cm) followed by L7 (-11.06 cm), L44 (-10.65 cm), L38 (-10.65 cm), L42 (-10.23 cm), L3, L12 and L48 (-9.81 cm). Inbred lines with highly significant negative GCA values for EPO were L3 and L45 (-0.06), L44 (-0.05), L42, L38, L48, L2, L46, L47, L12 (-0.04) and L7 (-0.03).

Inbred lines L6 (-7.32%) and L55 (-7.28%) were the only lines that had significantly negative GCA values for SL. Inbred lines with highly significant (<0.01) and negative GCA effects for CLR were L33 (-0.33) and L56 (-0.27); for EA were L45 (-0.53), L44 (-0.40), L32, L58 and L30 (-0.36) and for PA were L14, L35, L4, L41 and L3 (-0.25).

GCA effects of L19 (16.39 mm), L35 (16.08 mm), L21 (13.73 mm), L34 (9.92 mm), L36 (8.79 mm), L18 (8.59 mm) and L52 (8.44 mm) were positive and highly significant for EL. For KPR, L45 (3.42), L44 (2.27), L4 (2.22) and L35 (1.99) were the best inbred lines with highly significant and positive GCA effects. Inbred lines with highly significant and positive GCA effects for RPE were L31 (1.05), L47 (0.95), L38 (0.90), L45 (0.80), L25 (0.75), L48 (0.60), L2 (0.55) and L15 (0.55). For TKW, inbred lines L36 (26.43 g), L19 (21.20 g) and L5 (20.55 g) had highly significant and positive GCA effects followed by L57 (15.97 g), L10 (15.05 g), L53 (14.39 g) and L54 (14.12 g).

Table 13. Estimates of general combining ability (GCA) effects of lines and testers for grain yield and other agronomic traits in QPM hybrids

Lines	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L1	-0.22	-0.04	0.60	7.34	-0.65	-0.03 *	-6.80	0.17 *	0.39 **	0.12	-3.01	-1.19	0.45 *	-12.18
L2	0.38	-0.04	-0.65	7.34	-5.23	-0.04 **	-0.50	-0.02	-0.19	-0.19 *	2.64	0.97	0.55 **	8.66
L3	0.21	0.08	0.10	9.21 *	-9.81 **	-0.06 **	-5.39	0.04	0.14	-0.25 **	-0.95	-0.26	0.25	-0.02
L4	0.57 *	0.46	-0.40	6.09	1.02	0.01	-0.89	-0.02	-0.07	-0.25 **	2.71	2.22 **	-0.05	2.95
L5	0.09	1.08 *	0.73	-2.04	-3.56	-0.01	-5.30	0.04	-0.19	-0.07	-3.42	-1.68 *	-0.15	20.55 **
L6	-0.55 *	0.08	2.10 **	-5.79	-4.40	-0.01	-7.32 *	0.11	0.14	-0.13	-6.92 *	-1.26	-0.45 *	4.59
L7	-0.14	-0.67	2.10 **	-9.54 *	-11.06 **	-0.03 **	-5.13	0.17 *	-0.03	0.00	-3.57	-1.06	-0.15	4.75
L8	-0.39	0.58	0.98 *	-12.66 **	-7.31 *	-0.01	-5.35	0.04	0.10	-0.07	-5.54	-2.46 **	-0.40 *	2.81
L9	-0.35	-0.54	1.10 **	-3.91	-7.73 *	-0.02 *	-2.84	0.17 *	0.02	0.06	-6.46 *	-0.89	0.05	10.34
L10	-0.55 *	-1.04 *	2.35 **	-11.41 *	-8.56 *	-0.03 *	-2.11	0.23 **	-0.11	-0.07	-11.29 **	-2.81 **	0.45 *	15.05 *
L11	-0.61 *	0.21	1.23 **	-8.29	-4.81	0.00	-4.92	-0.02	0.14	-0.07	-7.26 *	-2.16 **	-0.20	-6.91
L12	0.03	-1.17 **	0.98 *	-8.29	-9.81 **	-0.04 **	-3.17	0.17 *	0.06	-0.13	-4.03	-1.18	0.35	7.90
L13	-0.17	1.58 **	1.35 **	4.84	3.52	0.01	-1.29	-0.08	-0.07	-0.19 *	-3.62	-0.36	0.30	-11.62
L14	-0.02	0.83 *	0.48	5.46	2.27	0.00	-1.64	0.17 *	-0.28 *	-0.25 **	-4.46	-0.31	-0.40 *	-6.53
L15	0.06	2.08 **	0.98 *	-1.41	-0.23	0.00	-6.01	0.11	-0.03	-0.13	-5.39	-0.21	0.55 **	-7.85
L16	0.13	-0.17	0.48	10.46 *	1.02	-0.02	-4.53	0.17 *	-0.19	-0.19 *	-6.18 *	-0.89	0.05	-4.25
L17	-0.33	-1.04 *	0.98 *	-10.79 *	-6.48 *	-0.01	-0.82	-0.08	0.10	-0.13	0.58	-0.09	-0.35	12.33
L18	0.20	0.46	-0.02	6.71	11.02 **	0.04 **	3.04	0.04	-0.19	0.06	8.59 **	0.51	0.05	-7.07
L19	0.16	1.21 **	0.23	13.59 **	9.35 **	0.01	0.61	0.17 *	0.06	0.12	16.39 **	-0.11	0.10	21.20 **
L20	0.13	-0.29	0.23	14.21 **	6.85 *	0.00	9.56 *	0.11	-0.19	0.06	7.59 *	0.02	-0.35	4.68
L21	0.56 *	-0.67	-0.77 *	16.09 **	13.52 **	0.03 *	4.19	0.17 *	-0.19	-0.07	13.73 **	0.64	0.33	3.90
L22	-0.20	-1.79 **	0.60	-3.29	-2.31	0.00	-0.20	-0.08	-0.07	-0.07	1.62	0.34	0.30	-2.83
L23	-0.23	-0.79 *	0.10	-1.41	10.60 **	0.06 **	25.01 **	-0.02	0.39 **	-0.07	-1.28	1.42 *	-0.15	-12.53
L24	-0.28	-0.92 *	0.10	-0.79	13.94 **	0.07 **	18.82 **	0.11	0.14	0.12	-6.65 *	-0.09	0.15	-16.94 *
L25	-0.40	1.58 **	-0.15	7.34	13.94 **	0.05 **	23.92 **	0.04	0.35 **	0.06	-0.11	-0.66	0.75 **	-25.81 **
L26	-0.48 *	0.33	0.85 *	1.71	6.02	0.03 *	13.66 **	-0.02	0.35 **	0.18 *	-11.16 **	-0.59	0.15	-10.15

Table 13. Estimates of general combining ability (GCA) effects continued

Lines	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L27	-0.53 *	-1.29 **	0.10	2.96	12.69 **	0.06 **	22.08 **	0.17 *	0.64 **	0.06	-5.16	-1.41 *	0.20	-21.82 **
L28	0.19	-0.67	-0.90 *	7.34	12.69 **	0.05 **	18.36 **	0.17 *	0.43 **	0.12	6.89 *	-0.16	0.00	-0.31
L29	-0.55 *	-0.17	-0.15	2.96	13.52 **	0.07 **	24.23 **	0.11	0.64 **	0.12	-2.54	0.12	-0.30	-24.40 **
L30	0.16	0.46	-0.77 *	1.09	-0.23	-0.01	-6.23	0.04	-0.36 **	-0.13	-1.94	0.39	0.10	-1.70
L31	0.37	-1.04 *	-0.40	0.46	-3.98	-0.02	-4.81	-0.21 *	-0.11	0.12	-4.04	1.49 *	1.05 **	10.60
L32	0.49 *	-0.17	-1.65 **	6.09	3.52	0.00	-2.96	-0.14	-0.36 **	0.18 *	2.17	0.99	-0.20	7.94
L33	-0.12	-0.04	-0.27	-1.41	-5.65	-0.02	-0.89	-0.33 **	0.22	0.06	2.99	1.34	-0.05	-0.69
L34	-0.06	1.33 **	-0.77 *	-2.66	5.60	0.02	-5.74	-0.02	0.27 *	0.06	9.92 **	0.14	-1.15 **	1.42
L35	0.83 **	0.21	-1.65 **	12.96 **	9.35 **	0.01	-3.92	-0.21 *	-0.15	-0.25 **	16.08 **	1.99 **	-0.05	12.34
L36	-0.05	-0.92 *	-1.65 **	12.34 *	6.85 *	0.01	-2.77	-0.08	-0.07	0.06	8.79 **	-0.14	-0.55 **	26.43 **
L37	0.33	1.33 **	-0.77 *	12.34 *	7.69 *	0.01	-2.96	-0.08	-0.28 *	-0.13	5.94 *	-0.04	-0.45 *	3.80
L38	0.06	-1.54 **	0.23	-8.29	-10.65 **	-0.04 **	0.29	0.11	0.27 *	0.12	0.63	1.24	0.90 **	-8.97
L39	0.15	-1.79 **	-0.40	1.09	2.27	0.01	-4.65	-0.02	-0.03	0.06	-0.72	0.29	-0.25	8.17
L40	-0.10	-0.17	-1.52 **	-6.41	5.60	0.04 **	-4.56	-0.21 *	0.06	0.12	-0.22	0.54	-0.80 **	10.00
L41	-0.32	0.71	0.85 *	-2.04	4.77	0.02 *	8.07 *	-0.02	-0.07	-0.25 **	-7.35 *	-2.69 **	0.35	-4.58
L42	-0.43	2.96 **	-0.77 *	-3.29	-10.23 **	-0.04 **	-5.88	0.04	-0.23 *	0.06	1.86	0.59	0.25	-17.12 *
L43	-0.29	1.08 *	0.60	-2.66	-8.15 *	-0.03 *	-3.24	-0.08	-0.11	0.18 *	1.63	0.56	0.15	-24.19 **
L44	-0.14	1.08 *	-0.27	-0.79	-10.65 **	-0.05 **	-4.58	0.04	-0.40 **	0.06	4.34	2.27 **	0.50 *	-26.31 **
L45	0.68 **	-1.42 **	0.73	-0.16	-13.15 **	-0.06 **	-6.81	-0.14	-0.53 **	0.00	5.58	3.42 **	0.80 **	1.09
L46	-0.04	2.33 **	1.85 **	8.59	-3.98	-0.04 **	-6.68	-0.08	-0.03	0.12	-0.93	1.52 *	0.15	-10.51
L47	0.04	1.08 *	0.98 *	2.34	-6.06	-0.04 **	0.41	-0.08	-0.28 *	0.18 *	-4.95	1.21	0.95 **	-1.69
L48	-0.03	0.71	0.23	-4.54	-9.81 **	-0.04 **	5.41	-0.02	0.10	0.00	-0.75	0.79	0.60 **	-16.65 *
L49	-0.16	0.46	-1.40 **	-7.66	-1.48	0.02	-5.68	-0.02	0.14	0.12	-5.44	-1.66 *	0.05	1.93
L50	0.18	-0.04	-1.52 **	-7.04	0.19	0.01	-4.73	-0.02	-0.03	0.06	5.42	1.29	-0.55 **	3.56
L51	-0.21	-1.29 **	-1.15 **	-3.91	-3.15	0.00	-6.43	-0.14	0.10	0.12	-2.95	-0.78	-0.65 **	3.20
L52	0.54 *	-1.17 **	-0.90 *	-1.41	-3.56	-0.01	-1.17	-0.08	-0.03	0.12	8.44 **	1.66 *	-0.15	8.34
L53	0.63 *	-1.04 *	0.35	-1.41	7.69 *	0.04 **	0.46	0.04	-0.03	0.06	7.41 *	-0.66	-0.05	14.39 *

Table 13. Estimates of general combining ability (GCA) effects continued

Lines	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L54	0.24	-0.92 *	-1.65 **	-0.79	-1.90	0.00	-4.63	-0.02	-0.11	0.00	1.15	-1.09	-0.40 *	14.12 *
L55	0.24	0.33	-0.77 *	-15.16 **	-2.31	0.02	-7.28 *	-0.14	-0.07	0.12	-1.42	-0.66	-0.50 *	8.37
L56	-0.19	-0.29	-1.27 **	-18.91 **	-5.23	0.01	-6.87	-0.27 **	0.27 *	0.00	-7.65 *	-0.56	-0.50 *	-0.39
L57	0.14	-0.17	-0.90 *	-10.16 *	0.19	0.03 *	-5.18	-0.08	-0.07	-0.13	0.58	1.07	-0.90 **	15.97 *
L58	0.35	-1.29 **	-1.15 **	-2.66	-3.56	0.00	-5.25	-0.08	-0.36 **	0.12	-6.33 *	-0.91	-0.95 **	12.61
Min	-0.61	-1.79	-1.65	-18.91	-13.15	-0.06	-7.32	-0.33	-0.53	-0.25	-11.29	-2.81	-1.15	-26.31
Max	0.83	2.96	2.35	16.09	13.94	0.07	25.01	0.23	0.64	0.18	16.39	3.42	1.05	26.43
SE	0.273	0.470	0.462	5.342	3.885	0.015	4.382	0.098	0.142	0.103	3.473	0.845	0.228	7.933
Testers														
CML144	-0.27 **	0.69 **	-0.22 **	-0.51 **	2.97 **	0.02 **	1.44 **	0.01 **	0.03	0.02	-0.34	0.12	0.06	-14.11 **
CML159	0.27 **	-0.69 **	0.22 **	0.51 **	-2.97 **	-0.02 **	-1.44 **	-0.01 **	-0.03	-0.02	0.34	-0.12	-0.06	14.11 **
SE	0.023	0.147	0.039	0.205	0.595	0.004	0.518	0.004	0.025	0.025	0.705	0.119	0.096	1.558

* *P*< 0.05; ** *P*< 0.01; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean.

5.3.2. Specific combining ability effects

SCA effects of different crosses for yield and different agronomic traits are presented in Table 14. Crosses L52/CML159 and L46/CML159 showed highly significant ($P < 0.01$) SCA effect for GY followed by L7/CML144, L33/CML144, L3/CML144 and L18/CML159 which had significant ($P < 0.05$) SCA effects.

Cross combinations L30/CML159, L38/CML159, L29/CML159, L22/CML159 and L56/CML159 exhibited negative and significant estimate of SCA effects for DA while L15/CML144, L7/CML144, L52/CML159, L13/CML159, L36/CML159 and L56/CML159 showed negative and significant SCA effects for ASI.

For PH, the lines L56 and L49 exhibited highly significant positive SCA effects when crossed to CML159 but highly significant negative values when crossed to CML144. The cross L52/CML159 was among the top seven crosses with significant and positive SCA values for PH. For EH, L38/CML159 had highly significant and negative SCA effects while other seven crosses showed significantly negative SCA values. Eleven crosses showed significantly negative SCA effects for EPO among which L38/CML159 and L4/CML159 showed the lowest SCA effect.

SCA effects of L24/CML159, L23/CML159, L28/CML159 and L41/CML159 were highly significant and negative for SL while that of L35/CML159, L11/CML144, L54/CML144 and L10/CML144 had highly significant and negative SCA effect for CLR.

The following crosses had highly significant and positive SCA effects for different yield components: L53/CML144 and L22/CML144 for KPR, L49/CML159, L47/CML144 and L31/CML144 for RPE and L19/CML144 for TKW. Crosses L43/CML159 and L53/CML144 had significant and positive SCA effects for EL.

Table 14. Estimates of specific combining ability (SCA) effects of line × tester for grain yield and other agronomic traits in QPM hybrids

Hybrids	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L1/CML144	0.39	0.44	-0.78	-0.12	-0.47	0.00	-0.70	0.11	-0.24	0.10	1.59	-0.32	0.34	7.71
L1/CML159	-0.39	-0.44	0.78	0.12	0.47	0.00	0.70	-0.11	0.24	-0.10	-1.59	0.32	-0.34	-7.71
L2/CML144	0.26	-0.31	-0.03	8.63 *	3.28	0.00	-5.20 *	-0.08	0.01	0.04	2.26	0.15	0.24	-3.91
L2/CML159	-0.26	0.31	0.03	-8.63 *	-3.28	0.00	5.20 *	0.08	-0.01	-0.04	-2.26	-0.15	-0.24	3.91
L3/CML144	0.51 *	-0.44	-0.78	8.01	1.19	-0.01	-1.05	-0.14	-0.32 *	-0.02	1.20	0.31	0.24	13.85 *
L3/CML159	-0.51 *	0.44	0.78	-8.01	-1.19	0.01	1.05	0.14	0.32 *	0.02	-1.20	-0.31	-0.24	-13.85 *
L4/CML144	0.38	0.44	-0.78	4.88	3.69	0.03 **	-1.99	-0.08	-0.28 *	0.10	5.64	-0.10	-0.26	13.62 *
L4/CML159	-0.38	-0.44	0.78	-4.88	-3.69	-0.03 **	1.99	0.08	0.28 *	-0.10	-5.64	0.10	0.26	-13.62 *
L5/CML144	-0.43	0.56	0.09	-5.74	-4.22	-0.01	-0.70	-0.01	0.01	-0.08	0.48	-0.37	-0.06	-3.42
L5/CML159	0.43	-0.56	-0.09	5.74	4.22	0.01	0.70	0.01	-0.01	0.08	-0.48	0.37	0.06	3.42
L6/CML144	0.34	-0.44	-0.03	-0.74	3.28	0.02	-2.25	-0.08	-0.15	-0.02	0.41	-1.15	-0.06	3.33
L6/CML159	-0.34	0.44	0.03	0.74	-3.28	-0.02	2.25	0.08	0.15	0.02	-0.41	1.15	0.06	-3.33
L7/CML144	0.52 *	-0.69	-1.28 **	10.51 *	7.44 *	0.02	-0.67	0.11	-0.32 *	-0.02	2.85	0.58	0.04	8.24
L7/CML159	-0.52 *	0.69	1.28 **	-10.51 *	-7.44 *	-0.02	0.67	-0.11	0.32 *	0.02	-2.85	-0.58	-0.04	-8.24
L8/CML144	-0.17	0.06	0.34	-6.37	-2.97	0.00	-1.14	-0.01	0.05	0.04	-0.52	-1.12	-0.01	-6.87
L8/CML159	0.17	-0.06	-0.34	6.37	2.97	0.00	1.14	0.01	-0.05	-0.04	0.52	1.12	0.01	6.87
L9/CML144	0.23	-0.31	-0.28	2.38	-0.89	-0.01	-2.42	-0.01	0.05	0.04	4.07	0.41	-0.46 *	16.12 *
L9/CML159	-0.23	0.31	0.28	-2.38	0.89	0.01	2.42	0.01	-0.05	-0.04	-4.07	-0.41	0.46 *	-16.12 *
L10/CML144	0.17	0.19	-0.78	3.63	-2.56	-0.02	3.05	-0.20 **	0.10	0.04	1.09	-0.17	0.04	3.41
L10/CML159	-0.17	-0.19	0.78	-3.63	2.56	0.02	-3.05	0.20 **	-0.10	-0.04	-1.09	0.17	-0.04	-3.41
L11/CML144	0.04	-0.31	0.09	-8.24	-1.31	0.01	-1.01	-0.20 **	0.10	0.04	0.90	-0.29	0.19	4.93
L11/CML159	-0.04	0.31	-0.09	8.24	1.31	-0.01	1.01	0.20 **	-0.10	-0.04	-0.90	0.29	-0.19	-4.93
L12/CML144	0.00	-0.69	-0.41	-5.74	-1.31	0.01	-3.32	0.11	0.01	0.10	-0.69	-1.30	-0.26	12.02
L12/CML159	0.00	0.69	0.41	5.74	1.31	-0.01	3.32	-0.11	-0.01	-0.10	0.69	1.30	0.26	-12.02
L13/CML144	-0.17	-0.69	0.97 *	-3.87	2.03	0.01	-0.16	-0.01	0.05	0.29 **	1.02	-0.72	-0.11	-1.00
L13/CML159	0.17	0.69	-0.97 *	3.87	-2.03	-0.01	0.16	0.01	-0.05	-0.29 **	-1.02	0.72	0.11	1.00

Table 14. Estimates of specific combining ability (SCA) effects continued

Hybrids	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L14/CML144	-0.01	-0.44	0.09	-0.74	2.44	0.01	3.32	-0.01	0.10	-0.02	1.05	0.70	0.19	-13.14
L14/CML159	0.01	0.44	-0.09	0.74	-2.44	-0.01	-3.32	0.01	-0.10	0.02	-1.05	-0.70	-0.19	13.14
L15/CML144	0.09	-0.44	-1.41 **	4.88	-0.06	-0.01	0.15	0.17 *	-0.07	0.10	0.36	0.10	0.04	9.96
L15/CML159	-0.09	0.44	1.41 **	-4.88	0.06	0.01	-0.15	-0.17 *	0.07	-0.10	-0.36	-0.10	-0.04	-9.96
L16/CML144	0.12	0.31	-0.16	4.26	5.36	0.01	-3.49	-0.14	0.18	-0.08	2.79	0.31	-0.06	-9.45
L16/CML159	-0.12	-0.31	0.16	-4.26	-5.36	-0.01	3.49	0.14	-0.18	0.08	-2.79	-0.31	0.06	9.45
L17/CML144	0.12	-0.56	0.59	-10.74 *	-2.14	0.01	4.67	0.11	-0.11	0.23 **	-4.84	-2.05 *	0.14	-4.12
L17/CML159	-0.12	0.56	-0.59	10.74 *	2.14	-0.01	-4.67	-0.11	0.11	-0.23 **	4.84	2.05 *	-0.14	4.12
L18/CML144	-0.46 *	0.19	0.09	-8.24	2.03	0.02	5.62 *	-0.01	0.35 *	0.17 *	3.46	0.31	0.04	4.27
L18/CML159	0.46 *	-0.19	-0.09	8.24	-2.03	-0.02	-5.62 *	0.01	-0.35 *	-0.17 *	-3.46	-0.31	-0.04	-4.27
L19/CML144	0.06	-0.56	-0.66	2.38	3.69	0.01	-0.32	0.11	-0.15	-0.02	-0.30	-0.10	-0.41	27.09 **
L19/CML159	-0.06	0.56	0.66	-2.38	-3.69	-0.01	0.32	-0.11	0.15	0.02	0.30	0.10	0.41	-27.09 **
L20/CML144	-0.38	0.44	-0.16	3.01	2.86	0.00	2.27	0.05	0.43 **	0.04	-0.04	-1.07	-0.06	-10.39
L20/CML159	0.38	-0.44	0.16	-3.01	-2.86	0.00	-2.27	-0.05	-0.43 **	-0.04	0.04	1.07	0.06	10.39
L21/CML144	0.14	-0.19	0.09	1.13	6.19 *	0.03 *	-0.73	0.11	-0.24	0.04	-3.30	0.38	-0.08	11.35
L21/CML159	-0.14	0.19	-0.09	-1.13	-6.19 *	-0.03 *	0.73	-0.11	0.24	-0.04	3.30	-0.38	0.08	-11.35
L22/CML144	0.20	0.94 *	0.22	-1.99	4.53	0.02	2.80	-0.01	-0.11	0.04	3.38	2.71 **	-0.11	9.92
L22/CML159	-0.20	-0.94 *	-0.22	1.99	-4.53	-0.02	-2.80	0.01	0.11	-0.04	-3.38	-2.71 **	0.11	-9.92
L23/CML144	0.17	0.69	-0.53	4.88	3.28	0.01	10.71 **	0.05	-0.07	0.04	2.28	0.20	0.14	-1.27
L23/CML159	-0.17	-0.69	0.53	-4.88	-3.28	-0.01	-10.71 **	-0.05	0.07	-0.04	-2.28	-0.20	-0.14	1.27
L24/CML144	-0.04	0.06	0.22	-0.74	-1.72	0.00	11.31 **	-0.08	0.01	-0.02	0.80	-0.79	-0.16	3.99
L24/CML159	0.04	-0.06	-0.22	0.74	1.72	0.00	-11.31 **	0.08	-0.01	0.02	-0.80	0.79	0.16	-3.99
L25/CML144	-0.17	0.56	0.22	-10.12 *	-3.39	0.00	-6.19 *	-0.01	0.05	0.04	0.39	0.95	0.14	0.45
L25/CML159	0.17	-0.56	-0.22	10.12 *	3.39	0.00	6.19 *	0.01	-0.05	-0.04	-0.39	-0.95	-0.14	-0.45
L26/CML144	-0.40	0.31	0.47	-4.49	-1.31	0.00	5.55 *	0.05	-0.03	0.04	-4.45	-0.99	0.04	-1.52
L26/CML159	0.40	-0.31	-0.47	4.49	1.31	0.00	-5.55 *	-0.05	0.03	-0.04	4.45	0.99	-0.04	1.52
L27/CML144	0.23	-0.06	0.22	-1.99	0.36	0.01	6.30 *	0.11	-0.07	0.04	5.14	0.63	0.09	0.05

Table 14. Estimates of specific combining ability (SCA) effects continued

Hybrids	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L27/CML159	-0.23	0.06	-0.22	1.99	-0.36	-0.01	-6.30 *	-0.11	0.07	-0.04	-5.14	-0.63	-0.09	-0.05
L28/CML144	0.12	-0.44	0.47	4.88	0.36	0.00	8.84 **	0.11	0.05	-0.02	0.95	-1.02	0.39	6.55
L28/CML159	-0.12	0.44	-0.47	-4.88	-0.36	0.00	-8.84 **	-0.11	-0.05	0.02	-0.95	1.02	-0.39	-6.55
L29/CML144	0.07	1.06 *	0.22	0.51	-3.81	-0.02	-0.81	0.05	-0.15	-0.15 *	3.25	0.63	-0.21	0.29
L29/CML159	-0.07	-1.06 *	-0.22	-0.51	3.81	0.02	0.81	-0.05	0.15	0.15 *	-3.25	-0.63	0.21	-0.29
L30/CML144	0.02	1.19 *	-0.16	6.13	-0.06	-0.01	-0.65	-0.01	-0.15	-0.02	1.76	0.96	-0.01	3.53
L30/CML159	-0.02	-1.19 *	0.16	-6.13	0.06	0.01	0.65	0.01	0.15	0.02	-1.76	-0.96	0.01	-3.53
L31/CML144	0.17	-0.06	-0.03	-0.74	0.36	0.00	-3.68	0.11	-0.24	0.10	4.81	1.03	0.74 **	9.72
L31/CML159	-0.17	0.06	0.03	0.74	-0.36	0.00	3.68	-0.11	0.24	-0.10	-4.81	-1.03	-0.74 **	-9.72
L32/CML144	0.32	0.56	-0.53	3.63	-1.31	-0.01	-3.33	0.05	-0.15	0.04	2.23	0.00	0.29	4.98
L32/CML159	-0.32	-0.56	0.53	-3.63	1.31	0.01	3.33	-0.05	0.15	-0.04	-2.23	0.00	-0.29	-4.98
L33/CML144	0.52 *	-0.06	0.59	-1.37	-4.64	-0.02 *	-2.54	-0.01	0.10	-0.08	3.03	1.55 *	-0.36	0.57
L33/CML159	-0.52 *	0.06	-0.59	1.37	4.64	0.02 *	2.54	0.01	-0.10	0.08	-3.03	-1.55 *	0.36	-0.57
L34/CML144	-0.13	0.31	0.59	-1.37	-1.72	-0.01	-1.12	-0.08	-0.03	-0.08	-2.92	-0.32	-0.06	-11.64
L34/CML159	0.13	-0.31	-0.59	1.37	1.72	0.01	1.12	0.08	0.03	0.08	2.92	0.32	0.06	11.64
L35/CML144	-0.18	0.19	-0.28	0.51	-1.31	-0.01	-0.92	0.24 **	-0.11	-0.02	-1.24	0.23	-0.06	-13.33
L35/CML159	0.18	-0.19	0.28	-0.51	1.31	0.01	0.92	-0.24 **	0.11	0.02	1.24	-0.23	0.06	13.33
L36/CML144	0.05	0.06	0.97 *	2.38	-2.97	-0.02	3.34	0.11	0.05	0.04	-2.52	-0.26	0.04	-17.52 *
L36/CML159	-0.05	-0.06	-0.97 *	-2.38	2.97	0.02	-3.34	-0.11	-0.05	-0.04	2.52	0.26	-0.04	17.52 *
L37/CML144	-0.01	-0.44	0.34	1.13	-4.64	-0.02	0.96	-0.01	-0.07	-0.02	-1.11	0.40	-0.06	0.38
L37/CML159	0.01	0.44	-0.34	-1.13	4.64	0.02	-0.96	0.01	0.07	0.02	1.11	-0.40	0.06	-0.38
L38/CML144	0.05	1.19 *	-0.16	5.51	10.36 **	0.04 **	-4.21	0.05	-0.20	-0.02	-0.62	-0.72	0.29	5.80
L38/CML159	-0.05	-1.19 *	0.16	-5.51	-10.36 **	-0.04 **	4.21	-0.05	0.20	0.02	0.62	0.72	-0.29	-5.80
L39/CML144	-0.10	-0.06	0.72	-2.62	0.78	0.01	-1.44	0.05	0.10	-0.21 **	-3.99	-1.70 *	0.24	-13.13
L39/CML159	0.10	0.06	-0.72	2.62	-0.78	-0.01	1.44	-0.05	-0.10	0.21 **	3.99	1.70 *	-0.24	13.13
L40/CML144	-0.12	-0.19	-0.16	-1.37	-5.89 *	-0.02 *	1.51	-0.01	0.01	-0.02	-2.95	-0.85	0.39	2.21
L40/CML159	0.12	0.19	0.16	1.37	5.89 *	0.02 *	-1.51	0.01	-0.01	0.02	2.95	0.85	-0.39	-2.21

Table 14. Estimates of specific combining ability (SCA) effects continued

Hybrids	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L41/CML144	-0.20	0.69	-0.78	-4.49	-5.89 *	-0.02	7.50 **	0.05	0.22	-0.02	-3.06	-0.92	-0.36	-6.85
L41/CML159	0.20	-0.69	0.78	4.49	5.89 *	0.02	-7.50 **	-0.05	-0.22	0.02	3.06	0.92	0.36	6.85
L42/CML144	0.25	-0.56	-0.66	8.01	-0.89	-0.02	-1.50	-0.01	-0.03	-0.08	-2.77	0.50	0.34	-7.92
L42/CML159	-0.25	0.56	0.66	-8.01	0.89	0.02	1.50	0.01	0.03	0.08	2.77	-0.50	-0.34	7.92
L43/CML144	-0.36	-0.69	0.22	1.13	-7.14 *	-0.03 *	-0.87	-0.01	0.01	-0.08	-7.83 *	0.03	-0.06	-9.87
L43/CML159	0.36	0.69	-0.22	-1.13	7.14 *	0.03 *	0.87	0.01	-0.01	0.08	7.83 *	-0.03	0.06	9.87
L44/CML144	-0.17	-0.19	0.09	0.51	-4.64	-0.03 *	-4.50	-0.01	-0.03	0.04	-3.08	0.45	0.39	5.56
L44/CML159	0.17	0.19	-0.09	-0.51	4.64	0.03 *	4.50	0.01	0.03	-0.04	3.08	-0.45	-0.39	-5.56
L45/CML144	-0.36	-0.69	0.59	-3.87	2.03	0.01	-1.70	-0.08	0.10	-0.02	0.83	0.40	-0.21	4.33
L45/CML159	0.36	0.69	-0.59	3.87	-2.03	-0.01	1.70	0.08	-0.10	0.02	-0.83	-0.40	0.21	-4.33
L46/CML144	-0.73 **	-0.19	0.72	-1.37	6.19 *	0.02 *	-2.89	-0.01	0.35 *	0.10	-5.04	-0.90	-0.26	-11.65
L46/CML159	0.73 **	0.19	-0.72	1.37	-6.19 *	-0.02 *	2.89	0.01	-0.35 *	-0.10	5.04	0.90	0.26	11.65
L47/CML144	0.24	0.06	-0.41	11.13 *	-1.72	-0.03 *	-4.75	-0.14	-0.24	-0.08	0.23	1.05	0.84 **	1.18
L47/CML159	-0.24	-0.06	0.41	-11.13 *	1.72	0.03 *	4.75	0.14	0.24	0.08	-0.23	-1.05	-0.84 **	-1.18
L48/CML144	0.03	-0.56	-0.16	10.51 *	5.36	0.01	-3.41	-0.08	-0.11	-0.15 *	-0.16	1.20	-0.41	5.27
L48/CML159	-0.03	0.56	0.16	-10.51 *	-5.36	-0.01	3.41	0.08	0.11	0.15 *	0.16	-1.20	0.41	-5.27
L49/CML144	-0.10	-0.06	-0.28	-12.62 **	-3.81	0.01	-0.15	-0.08	0.26 *	-0.02	0.15	-0.55	-0.96 **	-5.81
L49/CML159	0.10	0.06	0.28	12.62 **	3.81	-0.01	0.15	0.08	-0.26 *	0.02	-0.15	0.55	0.96 **	5.81
L50/CML144	0.06	0.69	0.09	5.51	3.69	0.01	-2.21	-0.08	-0.15	0.04	-1.33	0.76	-0.16	-1.32
L50/CML159	-0.06	-0.69	-0.09	-5.51	-3.69	-0.01	2.21	0.08	0.15	-0.04	1.33	-0.76	0.16	1.32
L51/CML144	-0.13	-0.81	-0.28	2.38	-0.47	-0.01	-1.53	0.17 *	0.22	-0.02	-3.20	-1.54 *	-0.26	-4.99
L51/CML159	0.13	0.81	0.28	-2.38	0.47	0.01	1.53	-0.17 *	-0.22	0.02	3.20	1.54 *	0.26	4.99
L52/CML144	-0.77 **	0.31	1.22 **	-10.12 *	2.44	0.03 *	4.39	-0.01	0.35 *	-0.02	-3.00	0.00	-0.46 *	-10.35
L52/CML159	0.77 **	-0.31	-1.22 **	10.12 *	-2.44	-0.03 *	-4.39	0.01	-0.35 *	0.02	3.00	0.00	0.46 *	10.35
L53/CML144	0.19	-0.56	-0.53	1.13	5.36	0.02	-2.02	-0.01	0.01	-0.08	7.09 *	2.95 **	0.14	0.33
L53/CML159	-0.19	0.56	0.53	-1.13	-5.36	-0.02	2.02	0.01	-0.01	0.08	-7.09 *	-2.95 **	-0.14	-0.33
L54/CML144	-0.14	0.56	0.22	0.51	-0.89	0.00	-0.90	-0.20 **	0.01	-0.15 *	0.39	0.01	-0.11	-9.15

Table 14. Estimates of specific combining ability (SCA) effects continued

Hybrids	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L54/CML159	0.14	-0.56	-0.22	-0.51	0.89	0.00	0.90	0.20 **	-0.01	0.15 *	-0.39	-0.01	0.11	9.15
L55/CML144	0.18	-0.69	0.09	4.88	-2.97	-0.03 *	-1.75	-0.08	-0.11	-0.02	0.73	-0.12	0.19	-0.53
L55/CML159	-0.18	0.69	-0.09	-4.88	2.97	0.03 *	1.75	0.08	0.11	0.02	-0.73	0.12	-0.19	0.53
L56/CML144	-0.12	0.94 *	0.84 *	-15.12 **	-7.56 *	-0.01	-1.26	0.05	0.14	-0.15 *	-5.01	-0.72	-0.01	-6.93
L56/CML159	0.12	-0.94 *	-0.84 *	15.12 **	7.56 *	0.01	1.26	-0.05	-0.14	0.15 *	5.01	0.72	0.01	6.93
L57/CML144	-0.30	0.06	0.22	-2.62	-2.14	-0.01	-1.10	-0.01	0.30 *	-0.15 *	-3.16	0.48	-0.31	-8.77
L57/CML159	0.30	-0.06	-0.22	2.62	2.14	0.01	1.10	0.01	-0.30 *	0.15 *	3.16	-0.48	0.31	8.77
L58/CML144	-0.11	-0.56	0.47	-1.37	-1.72	-0.01	-1.71	-0.14	0.18	-0.02	0.53	-0.24	0.14	-6.19
L58/CML159	0.11	0.56	-0.47	1.37	1.72	0.01	1.71	0.14	-0.18	0.02	-0.53	0.24	-0.14	6.19
SE	0.263	0.553	0.500	5.093	3.393	0.013	2.946	0.085	0.148	0.088	3.582	0.900	0.270	8.139

* *P*< 0.05; ** *P*< 0.01; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean.

5.3.3. Proportional contribution of GCA and SCA effects

Proportional contribution of line and tester GCA and line × tester SCA to the total variability among the hybrids are presented in Table 11 and Figure 1. The contribution of line GCA was found to be higher than the contribution of tester GCA and line × tester SCA for all considered traits except for TKW where the contribution of tester GCA was higher than that of line GCA and SCA. The contribution of the line × tester SCA was also higher than that of tester GCA for all traits under study except for DA and TKW. Line GCA contribution varied from 35.2% for TKW to 82.03% to SL; tester GCA's contribution ranged from 0.24% for EL to 47.02% for TKW while the SCA contribution ranged from 15.19% for DA to 36.64% for CLR.

5.4. Genotypic and phenotypic correlation

Genotypic and phenotypic correlation coefficients among grain yield and other agronomic traits across sites are presented in Table 15 while their biplot graph is presented in Figure 2. GY had positive and highly significant genotypic and phenotypic correlations with PH, EL and TKW but negative and highly significant correlation with DA, ASI, EPO, SL and EA. At genotypic level, GY had highly significant and negative association with CLR. At both genotypic and phenotypic level, SL was positively and highly significantly correlated with CLR, EA and PA. CLR had negative correlation with TKW at both genotypic and phenotypic levels, whereas it was negatively correlated with PA only at genotypic level. EH had positive and highly significant correlation with EPO, SL and EL but it had negative and highly significant correlation with TKW at both genotypic and phenotypic levels. EH also had positive and highly significant correlation with EA and PA only at genotypic level.

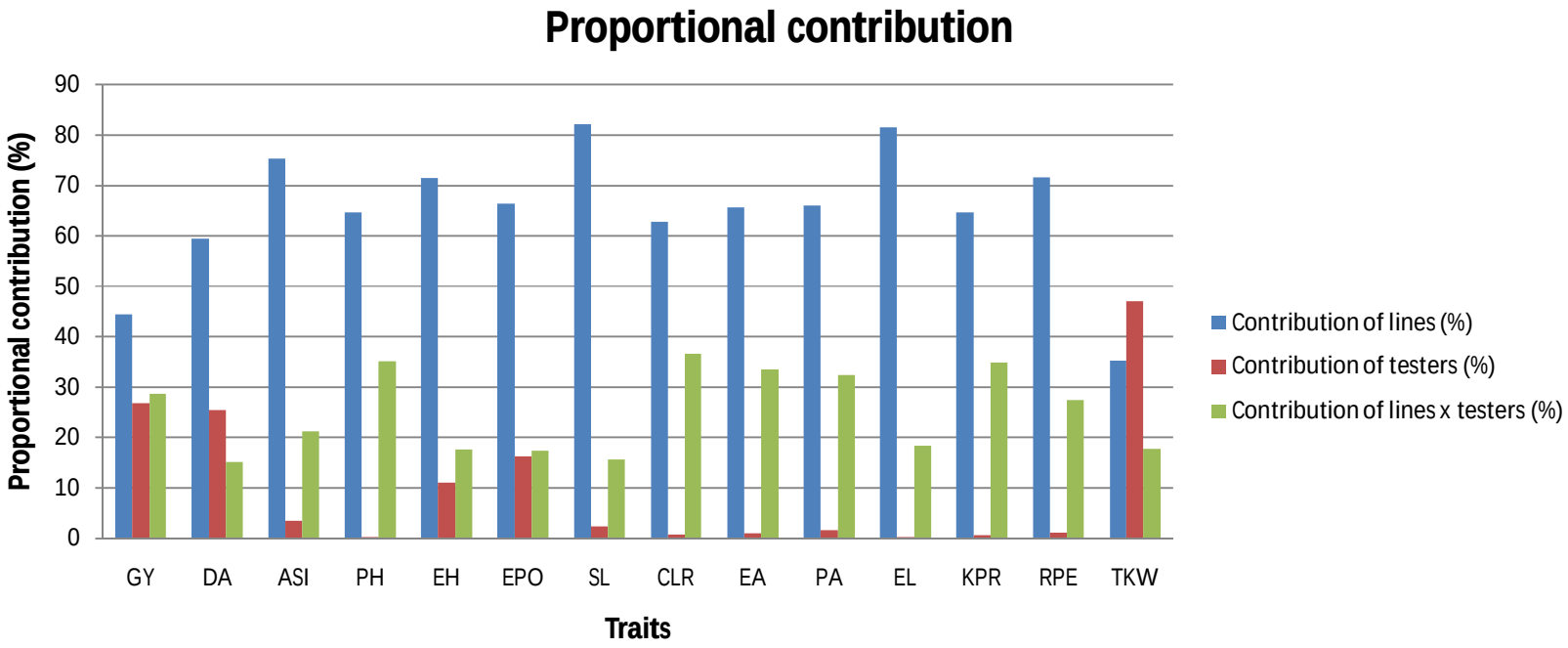


Figure 1. Proportional contribution of lines, testers and their interaction to the total hybrids variance (ASI= anthesis-silking interval; CLR= Common Leaf Rust (*Puccinia sorghi*); DA= days to anthesis; EA=ear aspect; EH= ear height; EL=ear length; EPO= ear position; GY= grain yield; KPR= number of kernels per row; PA=plant aspect; PH= plant height; RPE= number of rows per ear; SL=Shoot lodging;TKW= thousand kernel weight)

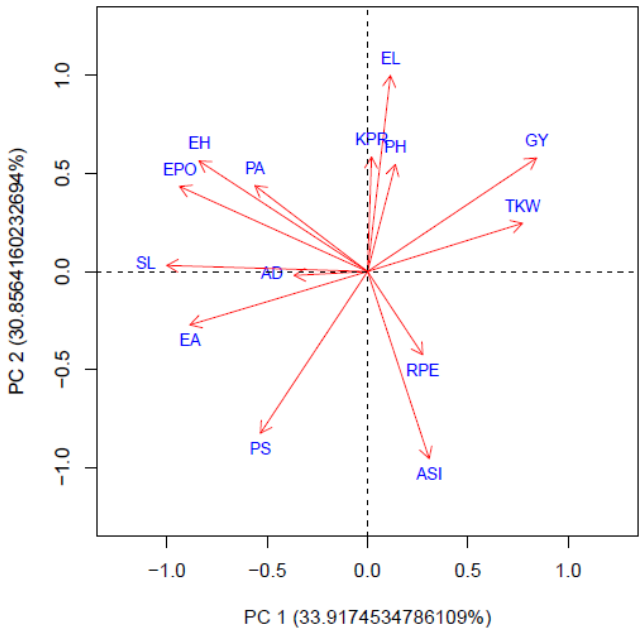


Figure 2. Biplot showing relationship among grain yield and other agronomic traits of QPM hybrids (ASI= anthesis-silking interval (d); PS= Common Leaf Rust (*Puccinia sorghi*) (1-5); AD= days to anthesis; EA=ear aspect (1-5); EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight)

EPO had highly significant and positive correlation with SL and EA but negative correlation with RPE and TKW both at phenotypic and genotypic levels. PH was significantly and positively correlated with EH, EL, KPR and RPE at both genotypic and phenotypic levels. DA had positive and highly significant correlation with PH and EH but negative correlation with TKW. At both genotypic and phenotypic levels, ASI was negatively and significantly correlated with PH, EH, EPO, PA, EL and KPR but positively correlated with CLR and RPE. CLR was positively and significantly correlated with EA and RPE but negatively correlated with EL at only genotypic level. However, it was correlated negatively and significantly with KPR at both genotypic and phenotypic levels. Both KPR and RPE were negatively and significantly correlated with TKW both at phenotypic and genotypic levels.

Table 15. Genotypic and phenotypic correlation among grain yield and other agronomic traits of QPM hybrids

Traits	DA		ASI		PH		EH		EPO		SL		CLR		EA		PA		EL		KPR		RPE		TKW	
GY	-0.39	**	-0.41	**	0.37	**	-0.18	*	-0.39	**	-0.32	**	-0.43	**	-0.50	**	-0.15		0.53	**	0.12		0.02		0.71	**
	-0.33	**	-0.29	**	0.30	**	-0.12		-0.26	**	-0.26	**	-0.22	*	-0.47	**	-0.17		0.42	**	0.19	*	-0.02		0.60	**
DA			-0.01		0.45	**	0.41	**	0.01		-0.17		0.12		-0.14		-0.18		-0.06		0.25	**	0.14		-0.73	**
			-0.05		0.26	**	0.28	**	0.18		0.04		-0.01		-0.03		-0.04		0.06		0.09		0.13		-0.55	**
ASI					-0.24	**	-0.63	**	-0.67	**	-0.20	*	0.41	**	0.01		-0.44	**	-0.60	**	-0.23	*	0.35	**	-0.08	
					-0.19	*	-0.37	**	-0.35	**	-0.04		0.20	*	0.11		-0.19	*	-0.46	**	-0.31	**	0.21	*	-0.04	
PH							0.62	**	0.00		0.00		0.00		-0.19	*	-0.32	**	0.58	**	0.53	**	0.39	**	-0.19	*
							0.46	**	0.06		0.13		0.07		-0.19	*	-0.14		0.45	**	0.26	**	0.20	*	-0.06	
EH									0.92	**	0.65	**	0.08		0.32	**	0.35	**	0.47	**	0.07		-0.21	*	-0.35	**
									0.90	**	0.49	**	0.05		0.20	*	0.09		0.33	**	0.05		-0.13		-0.24	**
EPO											0.67	**	-0.02		0.46	**	0.26	**	0.21	*	-0.06		-0.55	**	-0.33	**
											0.51	**	0.01		0.32	**	0.17		0.15		-0.04		-0.26	**	-0.25	**
SL													0.39	**	0.50	**	0.39	**	0.03		0.11		0.15		-0.48	**
													0.24	**	0.43	**	0.18		0.04		0.05		0.15		-0.35	**
CLR															0.49	**	0.00		-0.45	**	-0.30	**	0.35	**	-0.41	**
															0.06		0.05		-0.13		-0.24	**	0.13		-0.18	
EA																	0.08		-0.12		-0.19	*	-0.41	**	-0.20	*
																	0.07		-0.20	*	-0.19	*	-0.18		-0.26	**
PA																			0.21	*	0.54	**	-0.01		-0.15	
																			0.12		0.08		0.04		-0.07	
EL																					0.47	**	-0.24	**	0.14	
																					0.48	**	-0.11		0.17	
KPR																							0.31	**	-0.40	**
																							0.16		-0.20	*
RPE																									-0.45	**
																									-0.34	**

* *P*< 0.05; ** *P*< 0.01; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight; SE(m)=standard error of the mean.

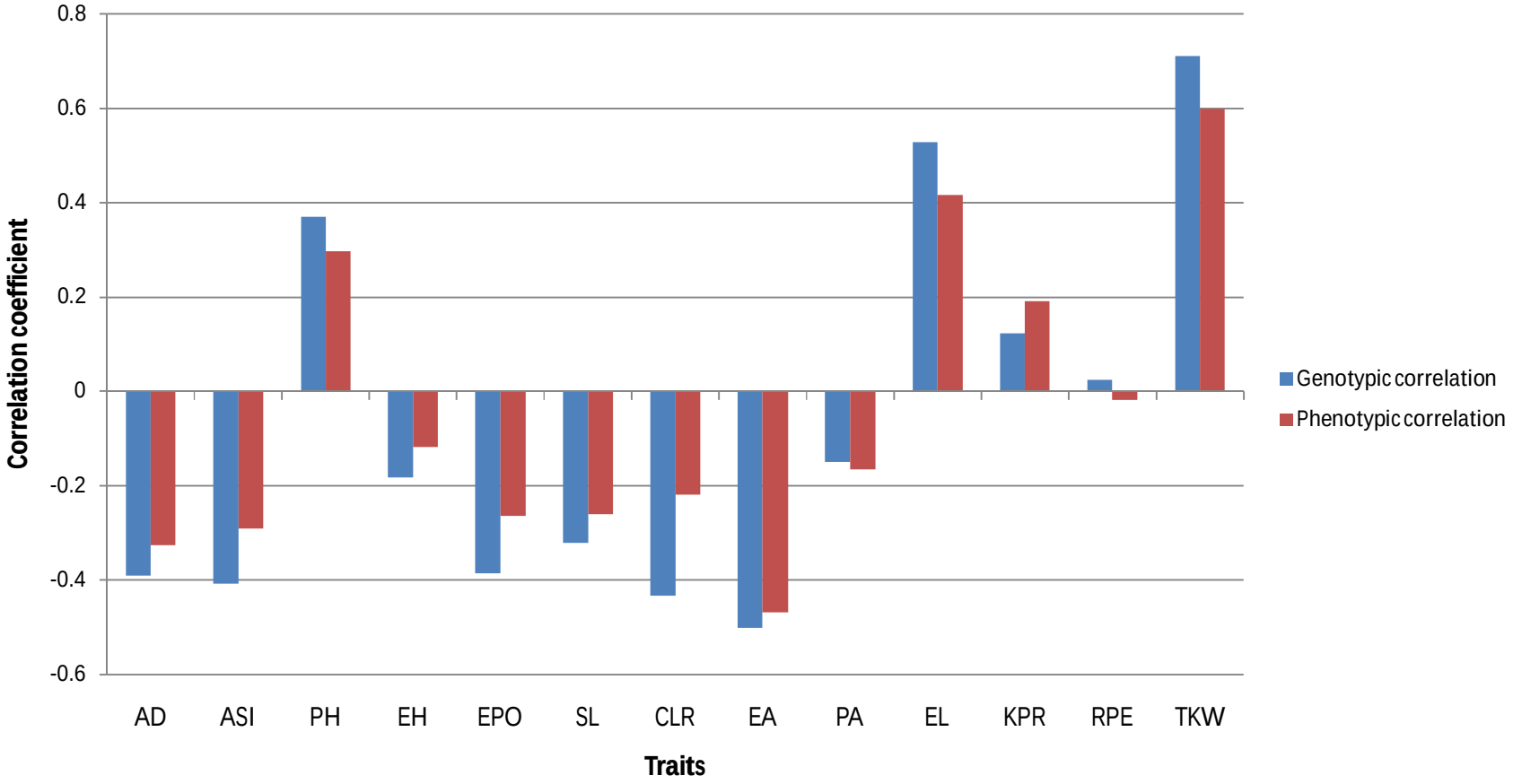


Figure 3. Genotypic and phenotypic correlations between grain yield and 13 other agronomic traits (ASI= anthesis-silking interval; CLR= Common Leaf Rust (*Puccinia sorghi*); DA= days to anthesis; EA=ear aspect ; EH= ear height; EL=ear length; EPO= ear position; KPR= number of kernels per row; PA=plant aspect; PH= plant height; RPE= number of rows per ear; SL=Shoot lodging;TKW= thousand kernel weight)

5.5. Components of variance, heritability and genetic advance

Estimates of genotypic, phenotypic and environmental variances of each site and across sites and genotype × site interaction variances are presented in Table 16. High genotypic, phenotypic and environmental variances were recorded for PH, EH, SL, EL and TKW at each site and across sites except the genotypic variance for SL at MARC. High genotype × site interaction variance was recorded only for SL and TKW. The highest environmental variances were recorded at Edo Gojola for all traits and the smallest was recorded at MARC.

Genotypic and phenotypic coefficients of variation, broad sense heritability, genetic advance and genetic advance as percentage of hybrid means estimates are presented in Table 17. High GCV and PCV values were recorded for ASI and SL while moderate PCV value was recorded for GY. All the other traits showed low PCV and GCV values. Very high heritability estimate was recorded only for DA whereas moderately high heritability values were recorded for ASI, EH, EPO, SL, EL, RPE and TKW. The traits that showed medium heritability estimates were GY, PH, CLR, EA and KPR while PA showed low heritability value. High percentage of GA was recorded for ASI and SL while GY, EH, EPO, EA and TKW exhibited moderate GA percentage. The percentage for all the other traits was low.

Table 16. Estimates of components of variances for grain yield and other agronomic traits of QPM hybrids

Trait	MARC			Edo Gojola			Mieso			Across			
	σ^2_e	σ^2_g	σ^2_p	σ^2_e	σ^2_g	σ^2_p	σ^2_e	σ^2_g	σ^2_p	σ^2_e	σ^2_g	σ^2_{gxe}	σ^2_p
GY	0.32	0.13	0.30	0.60	0.20	0.50	0.31	0.20	0.35	0.41	0.11	0.06	0.20
DA	1.21	1.53	2.14	1.55	2.20	2.98	-	-	-	1.40	1.79	0.07	2.17
ASI	1.24	0.83	1.45	1.28	1.18	1.82	-	-	-	1.26	0.88	0.14	1.27
PH	92.65	50.96	97.28	152.74	32.70	109.07	114.40	20.48	77.68	122.11	42.06	0.00	72.59
EH	87.72	69.42	113.29	147.53	96.19	169.96	103.85	30.53	82.45	113.18	57.45	8.13	79.03
EPO	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
RL	6.50	4.74	7.99	0.97	0.00	0.49	11.23	1.24	6.85	-	-	-	-
SL	20.69	8.51	18.85	181.85	176.85	267.78	78.99	55.72	95.21	94.47	53.43	28.44	78.66
EPP	0.01	0.01	0.02	0.01	0.00	0.01	0.02	0.00	0.01	-	-	-	-
CLR	0.04	0.01	0.03	0.06	0.03	0.06	0.09	0.00	0.05	0.05	0.01	0.01	0.03
EA	0.10	0.04	0.09	0.22	0.06	0.17	0.09	0.05	0.09	0.14	0.04	0.01	0.07
PA	0.04	0.02	0.04	0.05	0.01	0.03	0.06	0.02	0.05	0.05	0.01	0.01	0.02
ED	11.21	0.45	6.05	19.82	0.02	9.93	2.95	2.39	3.87	-	-	-	-
EL	65.25	37.20	69.83	86.64	32.57	75.88	69.54	27.11	61.88	73.80	24.75	7.00	39.38
KPR	3.82	1.54	3.45	4.15	1.57	3.64	5.03	1.51	4.02	4.30	0.99	0.58	1.90
RPE	0.40	0.18	0.38	0.46	0.06	0.29	0.44	0.26	0.48	0.42	0.20	0.02	0.31
TKW	299.24	373.04	522.66	571.31	348.36	634.02	509.60	385.40	640.20	464.48	313.88	47.02	406.96

σ^2_e = error variance; σ^2_g = genotypic variance; σ^2_p = phenotypic variance; σ^2_{gxe} = genotype × environment interaction variance; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); ED= ear diameter; EH= ear height (cm); EL=ear length; EPO= ear position; EPP= ears per plant; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RL= root lodging; RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight.

Table 17. Estimates of coefficients of variation, broad sense heritability and genetic advance for grain yield and other agronomic traits of QPM hybrids

Trait	MARC					Edo Gojola					Mieso					Across				
	GCV (%)	PCV (%)	H^2	GA	GA (% of $\bar{x}$)	GCV (%)	PCV (%)	H^2	GA	GA (% of $\bar{x}$)	GCV (%)	PCV (%)	H^2	GA	GA (% of $\bar{x}$)	GCV (%)	PCV (%)	H^2	GA	GA (% of $\bar{x}$)
GY	8.90	13.23	45.26	0.51	12.35	10.61	16.87	39.55	0.58	13.76	11.17	14.89	56.27	0.69	17.28	8.13	10.92	55.35	0.51	12.47
DA	1.78	2.10	71.72	2.16	3.11	1.97	2.29	73.94	2.63	3.50	-	-	-	-	-	1.85	2.04	82.34	2.50	3.46
ASI	182.51	240.97	57.37	1.43	285.18	60.46	75.04	64.91	1.81	100.49	-	-	-	-	-	81.72	97.95	69.60	1.62	140.64
PH	3.06	4.23	52.38	10.66	4.57	2.59	4.73	29.98	6.46	2.93	2.57	5.00	26.37	4.79	2.72	2.86	3.75	57.94	10.18	4.49
EH	7.00	8.94	61.28	13.46	11.30	8.60	11.43	56.60	15.22	13.34	7.01	11.53	37.03	6.94	8.81	7.29	8.55	72.70	13.33	12.82
EPO	6.24	7.98	61.23	0.05	10.08	7.51	9.77	59.18	0.06	11.92	7.73	11.24	47.23	0.05	10.96	6.97	7.99	76.11	0.06	12.54
RL	102.18	132.70	59.30	3.46	162.33	0.00	409.89	0.00	0.00	0.00	77.20	181.79	18.03	0.97	67.63	-	-	-	-	-
SL	109.25	162.63	45.13	4.04	151.41	76.38	93.99	66.04	22.30	128.06	176.05	230.13	58.52	11.78	277.84	90.24	109.49	67.93	12.43	153.44
EPP	9.88	12.45	62.98	0.17	16.18	3.75	9.01	17.35	0.03	3.22	3.32	10.69	9.64	0.02	2.13	-	-	-	-	-
CLR	3.94	6.68	34.70	0.12	4.78	7.60	10.38	53.52	0.28	11.46	0.00	10.64	0.00	0.00	0.00	4.82	7.04	46.93	0.17	6.82
EA	7.48	10.93	46.83	0.30	10.56	10.54	17.57	36.02	0.31	13.05	7.69	10.76	51.12	0.32	11.34	7.49	9.68	59.77	0.32	11.94
PA	5.24	7.71	46.10	0.18	7.34	3.93	7.71	26.01	0.10	4.14	6.04	9.55	40.05	0.18	7.89	3.20	6.49	24.33	0.08	3.26
ED	1.55	5.69	7.37	0.37	0.87	0.33	6.84	0.23	0.01	0.03	3.67	4.67	61.86	2.51	5.96	-	-	-	-	-
EL	4.01	5.49	53.28	9.18	6.04	3.76	5.75	42.92	7.71	5.09	3.57	5.39	43.81	7.11	4.87	3.32	4.19	62.84	8.14	5.43
KPR	3.51	5.25	44.60	1.71	4.83	3.67	5.59	43.08	1.70	4.97	3.77	6.17	37.44	1.55	4.76	2.93	4.06	52.17	1.49	4.37
RPE	2.86	4.14	47.56	0.60	4.06	1.60	3.60	19.68	0.22	1.46	3.64	4.96	53.91	0.77	5.52	3.08	3.87	63.56	0.73	5.07
TKW	9.88	11.70	71.37	33.66	17.22	9.55	12.89	54.95	28.54	14.61	8.70	11.22	60.20	31.42	13.93	8.62	9.82	77.13	32.10	15.62

GCV = Genotypic coefficient of variation; PCV = Phenotypic coefficient of variation; H^2 = Heritability; GA = Genetic advance; GA (% of $\bar{x}$) = Genetic advance as a percent of mean; ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); ED= ear diameter; EH= ear height (cm); EL=ear length; EPO= ear position; EPP= ears per plant; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RL= root lodging; RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight;

5.6. Heterosis

Mid-parent, better parent and standard heterosis of the hybrids among 58 inbred lines and two testers are summarized in Table 18. Analysis of BPH revealed that 49 out of 58 crossed involving CML144 and all of the crosses involving CML159 showed significantly positive heterosis for GY. Even though non-significant, five hybrids having CML144 as their parent and 19 hybrids having CML159 as their parent had positive heterosis over the standard check, MH140. Cross L39/CML159 showed the maximum BPH (327.6%) while L52/CML159 exhibited maximum SH with MH140 (19.7%) and MH130 (21.4%). In addition, hybrids L18/CML159 and L35/CML159 had standard heterosis more than 10% with both MH140 and MH130.

Significantly negative BPH for DA was observed in all the crosses. The lowest BPH was exhibited by L38/CML159 (-15.6%) followed by L22/CML159 (-15.4%). When compared to MH140, only three crosses involving the tester CML144 showed significant and negative BPH whereas 29 hybrids that involved CML159 showed significant and negative BPH for DA. However, all crosses showed positive SH as compared to MH130. Hybrids L38/CML159 and L22/CML159 had the smallest SH value for DA as compared to MH130.

BPH for PH and EH ranged from 24.70% (L17/CML144) to 68.84% (L53/CML159) and 23.58% (L33/CML144) to 97.28% (L32/CML159), respectively. All hybrids had significant and positive BPH for EH except L33/CML144, L44/CML144, L45/CML144, L56/CML144 and L43/CML159 which showed non-significant and positive heterosis. Non-significant SH was observed among the hybrids for PH when compared to MH140 while 21 hybrids had significant SH when compared to MH130. Conversely, none of the hybrids had significantly lower SH for EH when compared to MH130 while 13 hybrids had significantly negative SH as compared to MH140.

Almost all of the crosses showed significantly positive BPH for EL and KPR whereas none of the crosses exhibited significant and positive SH against MH140. Only L19/CML159 and L34/CML159 had significant and positive heterosis when compared to MH130. Hybrids L45/CML144 and L31/CML144 showed significantly positive standard heterosis for KPR when compared to MH140 while only L45/CML144 had significantly positive SH when compared to MH130.

Maximum SH for RPE was exhibited by L47/CML144 and L31/CML144 when compared to both MH130 and MH140 whereas 39 hybrids involving CML144 and 16 hybrids involving CML159 gave significantly positive BPH for the same trait.

Finally, the analysis of BPH showed that a total of 25 hybrids displayed significantly positive values for TKW with L4/CML144 displaying the maximum value (42.6%). The SH analysis revealed that there were non-significantly positive heterosis values for TKW.

Table 18. Minimum, maximum and mean heterosis over mid-parent, better parent and best checks for grain yield and other agronomic traits of QPM hybrids

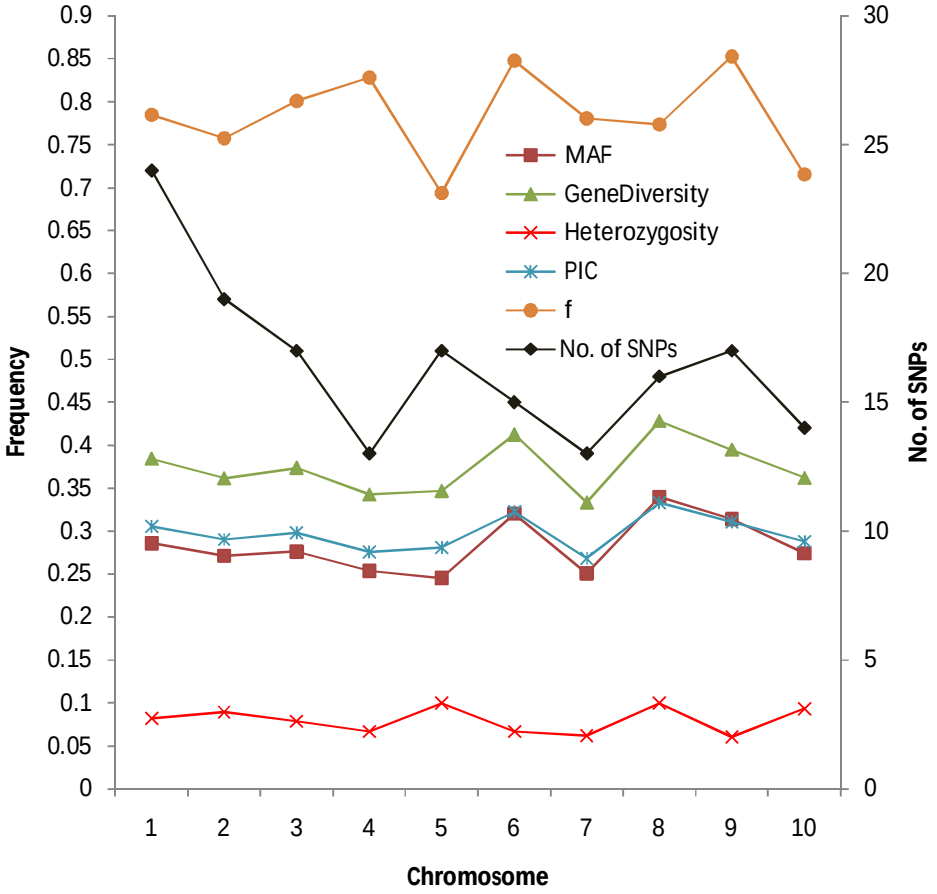
Traits	Mid-parent heterosis (%)			Better parent heterosis (%)			Standard heterosis (%)					
							MH140			MH130		
	Minimum	Maximum	Mean	Minimum	Maximum	Mean	Minimum	Maximum	Mean	Minimum	Maximum	Mean
GY	37.53	437.47	189.89	15.26	327.55	149.56	-32.00	19.67	-9.20	-31.03	21.38	-7.90
DA	-1.60	-10.24	-6.34	-7.70	-15.59	-12.28	-7.09	1.77	-1.92	2.81	12.61	8.53
ASI	-1238.49	122221.13	1125.78	-930.75	4036.49	-17.82	-167.60	182.77	-31.88	-205.13	339.77	5.94
PH	28.08	73.94	56.21	24.70	68.84	48.90	-8.26	9.01	0.79	-3.12	15.11	6.43
EH	26.06	112.32	68.57	23.58	97.28	54.40	-29.71	8.18	-9.58	-4.13	47.56	23.33
EPO	-7.71	24.96	6.60	-13.72	16.72	1.19	-26.59	8.89	-7.56	-9.14	34.77	14.40
SL	-112.53	1394.37	56.46	-111.46	675.82	0.32	-108.72	1270.48	161.24	-103.43	439.47	2.83
CLR	-14.46	23.30	3.87	-28.08	19.41	-8.02	22.75	78.08	51.79	-20.81	14.88	-2.07
EA	-43.30	4.60	-20.62	-4.69	-48.43	-26.98	-22.70	27.46	0.86	-13.23	43.08	13.22
PA	-2.86	-31.63	-18.51	-7.80	-37.46	-22.49	-11.41	27.01	12.68	-24.76	7.88	-4.30
EL	15.42	51.08	30.92	11.56	37.50	21.98	-11.87	8.13	-3.60	-7.96	12.93	0.68
KPR	13.30	85.20	47.65	8.99	68.00	36.90	-5.48	16.17	4.78	-8.05	13.01	1.92
RPE	1.33	31.11	11.63	-4.84	28.83	8.90	-8.97	13.70	-0.27	-10.04	12.35	-1.45
TKW	2.83	56.58	24.81	-15.16	42.63	14.27	-33.92	8.54	-14.10	-35.69	5.64	-16.39

ASI= anthesis-silking interval (d); CLR= Common Leaf Rust (*Puccinia sorghi*) (1-5); DA= days to anthesis; EA=ear aspect (1-5); EH= ear height (cm); EL=ear length; EPO= ear position; GY= grain yield (t ha^{-1}); KPR= number of kernels per row; PA=plant aspect; PH= plant height (cm); RPE= number of rows per ear; SL=Shoot lodging (%);TKW= thousand kernel weight;

5.7. SNP analysis

5.7.1. SNP characteristics

Out of the 201 SNPs used in the KASP assay, 195 were successfully called in the 59 inbred lines. SNP markers that had missing data points more than 20% (13 SNPs) or minor allele frequency (MAF) of less than 5% (17 SNPs) were excluded from data analysis. As a result, a total of 165 high quality SNPs were used for further analysis. Number of SNPs, minor allele frequency, gene diversity, heterozygosity, polymorphic information content and inbreeding coefficient per chromosome for the selected 165 SNPs are summarized in Figure 4.



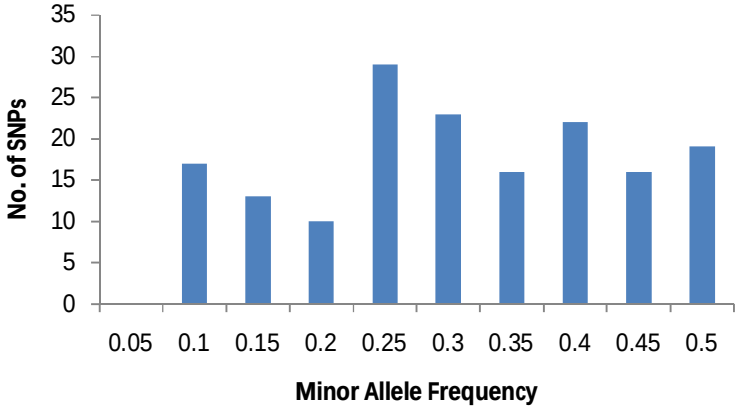
MAF=Minor allele frequency, PIC=Polymorphic information content, f=inbreeding coefficient

Figure 4. Summary statistics of the 165 SNPs used to characterize 59 QPM inbred lines. All the statistics are average of all SNPs per chromosome.

The selected SNPs were well distributed across the 10 chromosomes with an average of 17 SNPs per chromosome ranging from 24 SNPs on chromosome 1 to 13 SNPs on chromosome 4 and 7. Base changes involved A/C (32), A/G (77), A/T (14), C/G (16), C/T (21) and G/T (5) in which A/G transition was abundant accounting for 46.7% while G/T transversion was very rare accounting for only 3% of considered SNPs. A list of these high quality and informative SNPs with their chromosome, chromosome position, base change, flanking sequence and the gene locus where they are found is presented in Appendix 8.

Of the 165 SNP markers, 75.8% (125 of 165) had MAF more than 0.2 and 77.6% (128 of 165) had high PIC value which is >0.25. MAF close to 0.5 which shows almost equal allele frequencies for their alternative alleles was recorded at 19 (11.5%) SNP loci. The mean of MAF was 0.284 with a peak frequency between 0.2 and 0.25. PIC values ranged from 0.086 to 0.375 with a mean of 0.298 and a peak distribution between 0.35 and 0.4 (Figure 5). Heterozygosity ranged from 0.000 to 0.273 with average of 0.081. On the other hand, the average inbreeding coefficient was 0.789 ranging from 0.379 to 1.000. The range of gene diversity was 0.090 – 0.500 having a mean of 0.376.

(a)



(b)

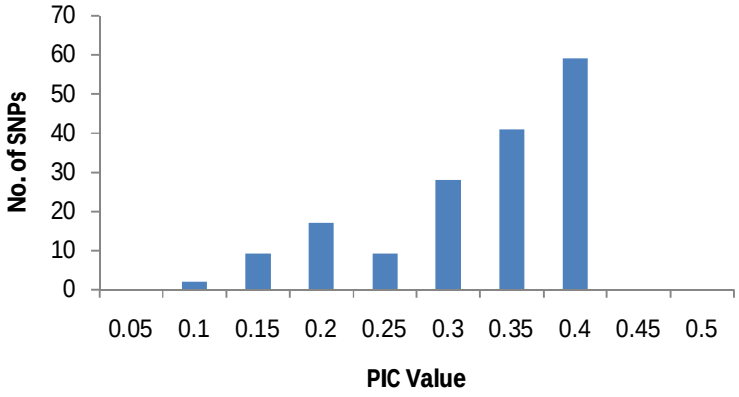


Figure 5. Frequency distribution of (a) minor allele frequency (MAF) and (b) polymorphic information content (PIC) among 59 inbred lines of maize

5.7.2. Population structure

To infer population structure for all the 59 inbred lines, the estimated Ln probability of the data (LnP(D)) using STRUCTURE and the computed ad hoc statistic ΔK using Evanno method are presented using graph in Figure 6. The Ln probability (LnP(D)) sharply increased from K=1 upto K=6 and started to reduce at K=7 and plateaued afterwards which seems to indicate that the best likely number of populations is six or seven. Similarly, the ad hoc statistic ΔK showed a peak at K=6. However, the ΔK also decreased when K increased from 7 to 8 as sharply as from 6 to 7 and at K=7, ΔK was much higher than at K=8. The slope became approximately zero after K=8. Therefore, K=6 and K=7 were considered as two best possible number of populations which supports the choice based on the plot of LnP(D) whereas K=7 matches with the population determined based on *a priori* pedigree knowledge. Therefore, the 59 inbred lines can be claimed to be clustered into seven distinct populations.

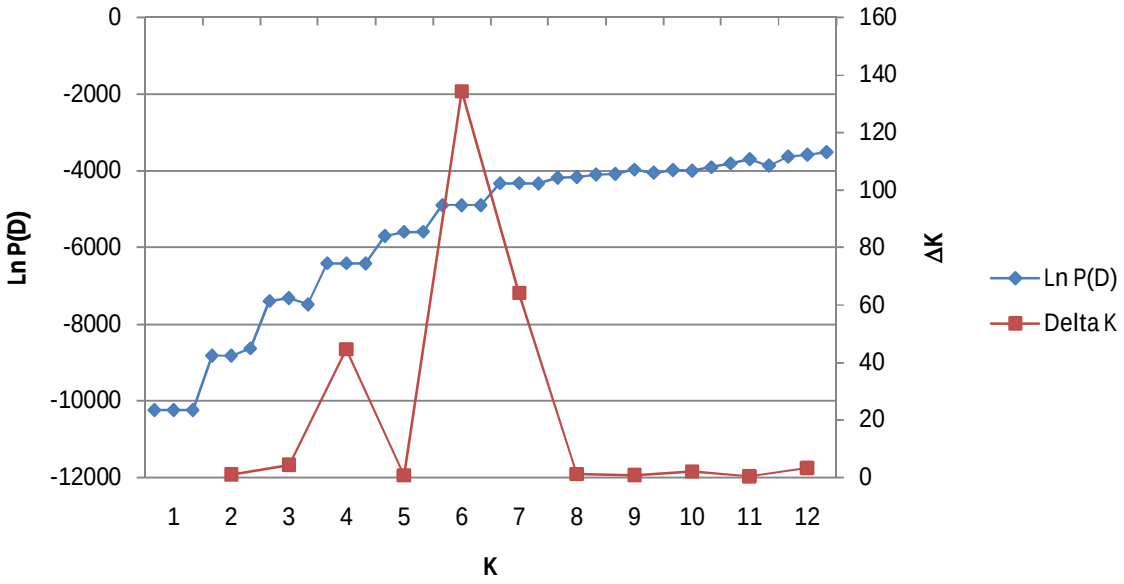


Figure 6. Plot of estimated Ln probability of the data (LnP(D)) and Evanno's ΔK for K=1-12 and over three K repeats

Among the 59 inbred lines, 47 were clearly assigned to distinct populations with probability ≥ 60 (Figure 7). Population 1, named ECA-EE-9, included all four ECA-EE-9 derived lines. The largest population according to pedigree information which included 17 inbred lines, Pop G, was subdivided into two distinct populations, population 2 named ECA-EE-34a and population 3 named ECA-EE-34b. The former included eight of them and the latter five of them. Population 4 included seven ECA-EE-13 derived lines and so named ECA-EE-13. The majority of the lines were assigned to population 5, ECA-EE-8, which included 13 ECA-EE-8 family. Population 6 included seven ECA-EE-16 derived lines and so called ECA-EE-16. The last one, population 7, included three ECA-EE-6 derived lines and so called ECA-EE-6. Two inbred lines were misplaced as ECA-EE-31/33 or Pop F and ECA-EE-16 or Pop E based on pedigree information and the former was assigned to population 3 or ECA-EE-34a with 78.3% probability while the latter to population 5 or ECA-EE-8 with 99.6% probability (Figure 7).

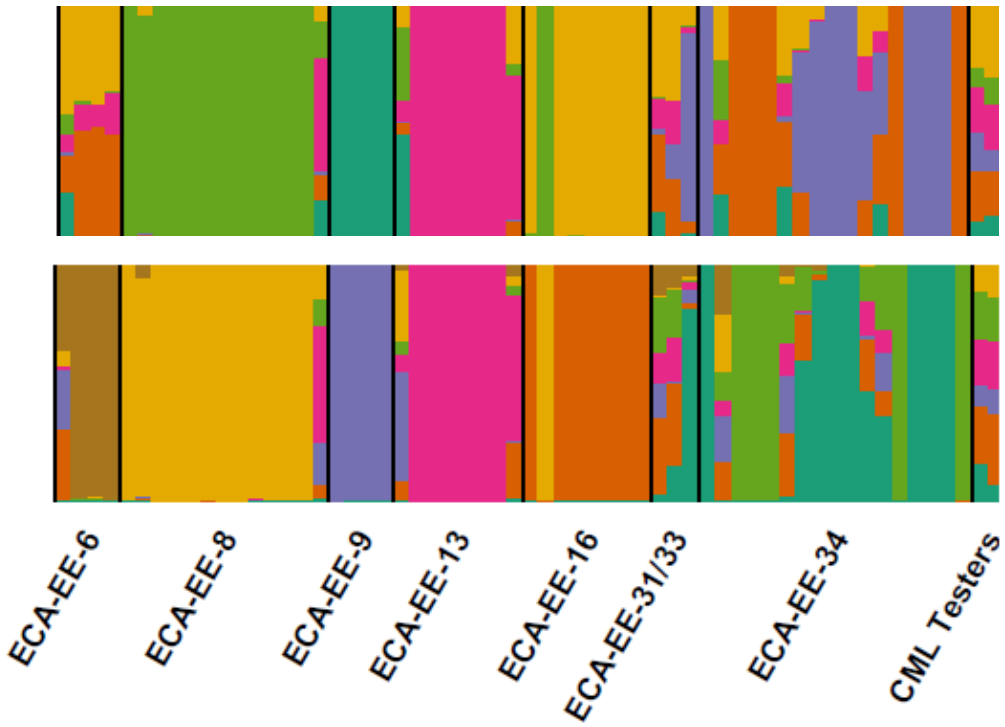


Figure 7. Population structure of 59 inbred lines genotyped with 165 SNPs assessed by STRUCTURE at K=6 (top) and K=7 (bottom)

The remaining 12 inbred lines could not be clearly assigned to any of these seven populations as they had membership probabilities < 60%. These lines, therefore, were classified into a mixed population. It included all the one line from ECA-EE-6, one line from ECA-EE-8, one line from ECA-EE-13, two lines from ECA-EE-31/33 and five lines from ECA-EE-34. In addition, both testers, CML144 and CML159 belonging to heterotic group B and A, respectively, based on conventional breeding approaches were assigned to the mixed population.

5.7.3. Cluster analysis

Roger's genetic distance between pairwise comparisons showed that 37% of the distances were between 0.4 and 0.45. The distance ranged from 0.000 to 0.561 while the average

distance was 0.377. Figure 8 shows percentage of Roger's genetic distance for the 59 inbred lines genotyped with 165 SNPs. Based on this distance, a neighbor-joining (NJ) tree was

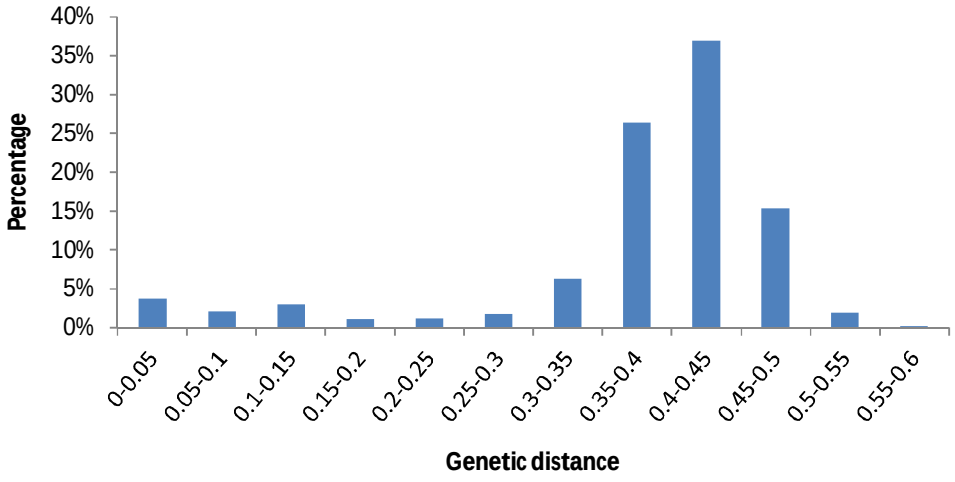


Figure 8. Percentage of Roger's genetic distance for 59 inbred lines genotyped with 165 SNPs

constructed which is presented in Figure 9. The populations identified by NJ tree totally matched with that of STRUCTURE at K=7. The 59 inbred lines were clustered into three major populations. The first population had three inbred lines that matched with STRUCTURE's ECA-EE-6 and the second population consisted of four inbred lines that was also consistent with ECA-EE-9 of STRUCTURE. The majority (40 inbred lines) fell into the third population which was divided into five subpopulations. These subpopulations were named Pop3a, Pop3b, Pop3c, Pop3d and Pop3e which were consistent with ECA-EE-34a, ECA-EE-34b, ECA-EE-13, ECA-EE-16 and ECA-EE-8 respectively and were constituted from 5, 8, 7, 7 and 13 inbred lines. The mixed population that was identified by STRUCTURE involving 12 inbred lines could also be clearly identified from cluster analysis with the same inbred lines (Figure 9).

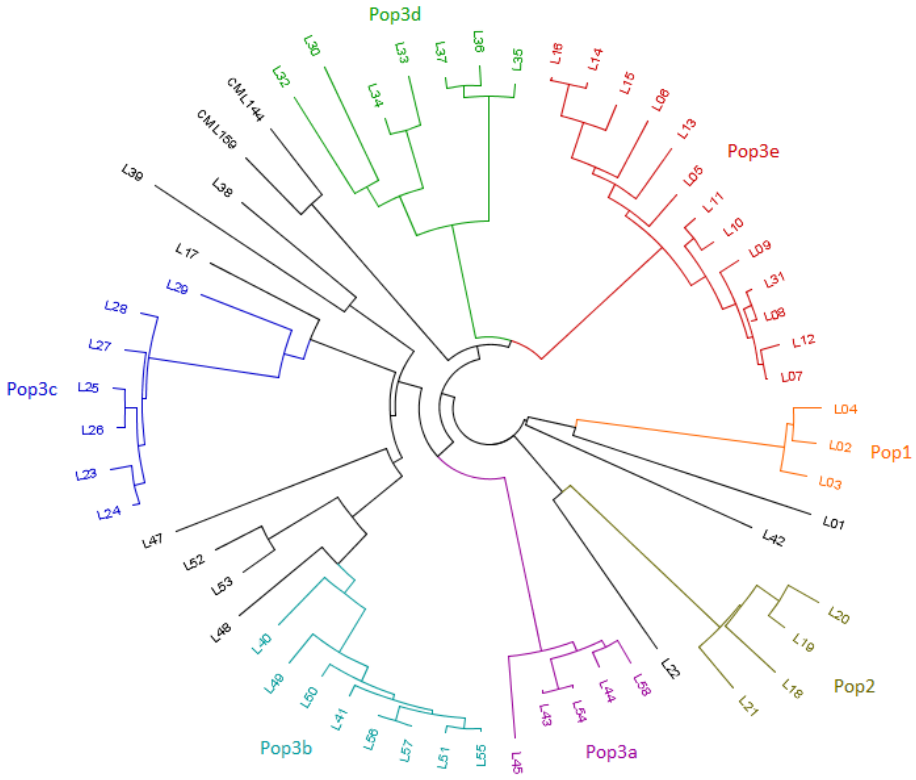


Figure 9. Neighbor-joining tree for 59 inbred lines based on Roger's genetic distance genotyped with 165 SNPs (Orange = ECA-EE-6, Olive green = ECA-EE-9, Purple = ECA-EE-34a, Aqua = ECA-EE-34b, Blue = ECA-EE-13, Green = ECA-EE-16, Red = ECA-EE-8, Black = mixed)

5.7.4. Principal component analysis

The first two principal components explained 36.27% of the total SNP variations among samples. A plot of PC1 (18.8%) and PC2 (17.47%) revealed seven major groups and a mixed group (Figure 10) that was more or less similar to STRUCTURE and cluster analysis. The difference was only that the population which included all the three inbred lines of ECA-EE-6 based on STRUCTURE and cluster analysis, included also the CML testers (CML144 and CML159) and three other lines. As a result, the mixed population was left with six inbred lines.

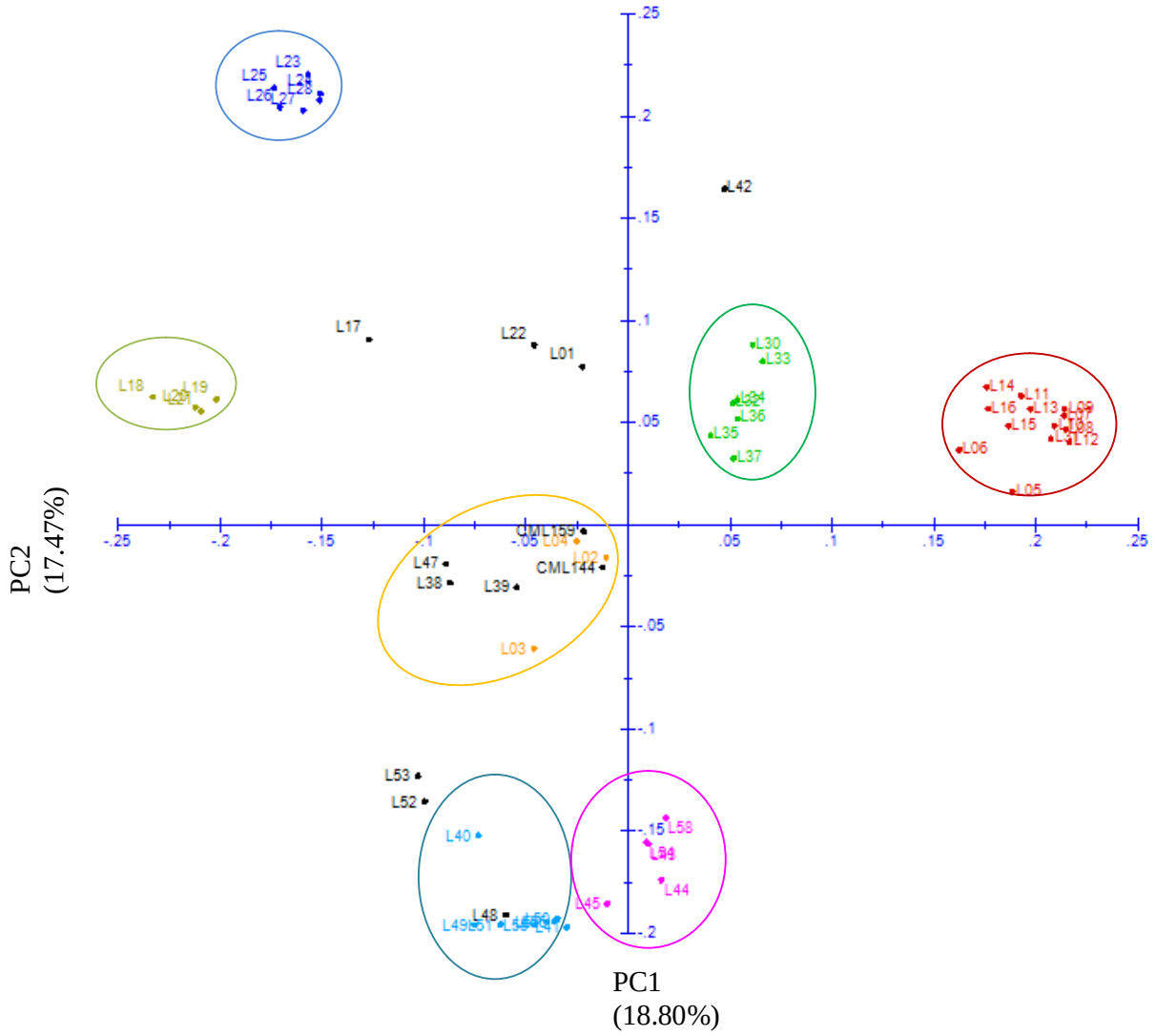


Figure 10. Principal component analysis of 59 inbred lines genotyped with 165 SNPs (Orange = ECA-EE-6, Olive green = ECA-EE-9, Purple = ECA-EE-34a, Aqua = ECA-EE-34b, Blue = ECA-EE-13, Green = ECA-EE-16, Red = ECA-EE-8, Black = mixed)

5.7.5. Analysis of molecular variance

Analysis of molecular variance (AMOVA) found that among the total variation, the highest variation (51.5%) was attributed to among populations variation while 29.3% was attributed to variation among individuals within populations (Table 19). Within individuals, there was 19.2% variation.

Table 19. Locus-by-locus analysis of molecular variance (AMOVA) for eight populations based on 165 SNPs

Source of variation	Sum of squares	Variance components	Percentage variation	Fixation indices	*P*
Among populations	1516.422	14.93862	51.49668	FST=0.51497	<0.00001
Among individuals within populations	1071.149	8.51328	29.34714	FIS=0.60505	<0.00001
Within individuals	309.000	5.55700	19.15618	FIT=0.80844	<0.00001
Total	2896.571	29.00890			

5.8. ISSR Analysis

5.8.1. ISSR primers and their polymorphism

Out of the 24 ISSR primers tested, seven (five di-, one tetra- and one penta-nucleotide) primers were selected that produced distinct amplified fragments and out of 58 samples, 35 (60%) yielded amplified products for all the selected seven primers. The number of polymorphic bands ranged from 10 bands for the penta-nucleotide primer 880 to 18 bands for the di-nucleotide primer 812 with a mean of 12.6 bands per primer. Using the selected primers, a total of 88 bands were scored and out of them 86 bands (97.73%) were found to be polymorphic. All the primers were 100% polymorphic except 812 (94.44%) and 835 (90.91%) (Table 20). The amplified fragments generated by primer 835 are presented in Figure 11 as an illustration.

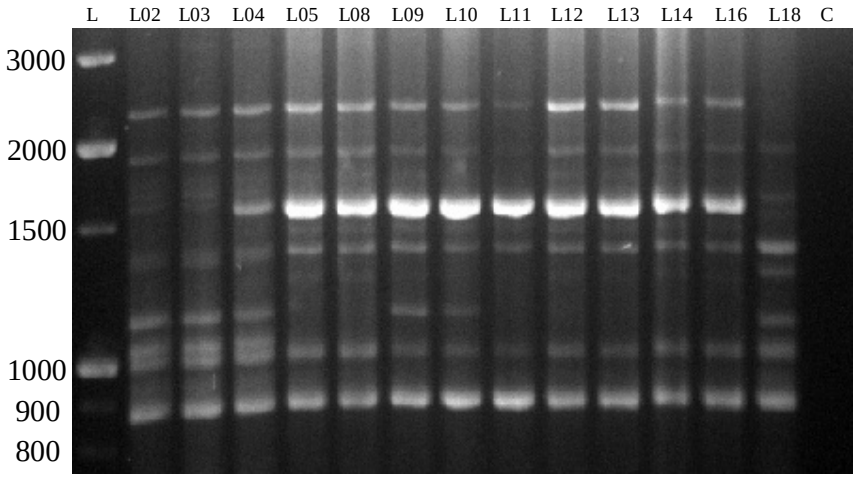


Figure 11. ISSR amplification profile for the first batch of maize inbred lines with primer 835 (L = DNA ladder; C = non template control).

Each of the seven primers revealed polymorphism within each of the populations except primers 835 and 880 in ECA-EE-6. This population exhibited the smallest percentage of polymorphism by the rest of the primers too. The highest percent polymorphism (100%) was produced by primer 880 in ECA-EE-8. The percent polymorphism overall the primers ranged from 5.68% in ECA-EE-6 to 69.32% in ECA-EE-16.

Table 20. Primers selected for ISSR analysis of 35 maize inbred lines and the number of loci and percent polymorphism they produced in each of the populations and across the populations clustered based on pedigree information

Primers	No. of loci	Percent polymorphism						
		ECA-EE-6	ECA-EE-8	ECA-EE-9	ECA-EE-13	ECA-EE-16	ECA-EE-30s	Total
812	18	5.56	50.00	27.78	38.89	83.33	61.11	94.44
824	11	9.09	63.64	54.55	63.64	63.64	81.82	100.00
835	11	0.00	36.36	27.27	54.55	63.64	36.36	90.91
836	14	7.14	78.57	50.00	78.57	71.43	64.29	100.00
848	13	7.69	38.46	15.38	23.08	38.46	76.92	100.00
873	11	9.09	72.73	18.18	27.27	72.73	63.64	100.00
880	10	0.00	100.00	80.00	70.00	90.00	80.00	100.00
Total	88	5.68	61.36	37.50	50.00	69.32	65.91	97.73

5.8.2. Genetic diversity analysis

Primer 880 and 835 showed the minimum (0.00) Nei's gene diversity in population ECA-EE-6 while primer 880 exhibited the highest (0.45) gene diversity in population ECA-EE-8 which is in agreement with their percent polymorphism. Overall, however, primer 836 revealed the highest diversity with a value of 0.42 and populations ECA-EE-16 and ECA-EE-34 with a value of 0.25 (Table 21). The lowest Nei's diversity was observed in population ECA-EE-6 (0.02) and primer 835 (0.31). Similar trends were observed using Shannon's information index across the primers and populations (Table 22).

Table 21. Nei's gene diversity (h) for the primers and populations

Primers	ECA-EE-6	ECA-EE-8	ECA-EE-9	ECA-EE-13	ECA-EE-16	ECA-EE-30s	Overall
812	0.02±0.073	0.15±0.196	0.10±0.180	0.08±0.110	0.30±0.184	0.22±0.214	0.35±0.143
824	0.04±0.113	0.15±0.179	0.18±0.181	0.21±0.197	0.27±0.211	0.22±0.181	0.35±0.114
835	0.00±0.000	0.10±0.171	0.10±0.176	0.21±0.221	0.21±0.208	0.12±0.193	0.31±0.145
836	0.02±0.080	0.33±0.198	0.20±0.222	0.29±0.201	0.28±0.209	0.23±0.218	0.42±0.078
848	0.04±0.135	0.13±0.195	0.06±0.153	0.11±0.214	0.15±0.213	0.30±0.199	0.32±0.171
873	0.05±0.122	0.31±0.203	0.07±0.153	0.13±0.230	0.27±0.195	0.27±0.224	0.36±0.149
880	0.00±0.000	0.45±0.049	0.34±0.192	0.25±0.209	0.25±0.144	0.39±0.205	0.41±0.151
Overall	0.02±0.086	0.23±0.210	0.15±0.196	0.18±0.203	0.25±0.196	0.25±0.212	0.37±0.124

Table 22. Shannon's information index (I) for the primers and populations

Primers	ECA-EE-6	ECA-EE-8	ECA-EE-9	ECA-EE-13	ECA-EE-16	ECA-EE-30s	Overall
812	0.03±0.116	0.23±0.279	0.16±0.263	0.14±0.184	0.45±0.250	0.32±0.299	0.52±0.186
824	0.07±0.180	0.24±0.253	0.28±0.273	0.32±0.282	0.39±0.296	0.35±0.241	0.53±0.133
835	0.00±0.000	0.16±0.252	0.15±0.260	0.31±0.314	0.31±0.291	0.18±0.274	0.47±0.194
836	0.03±0.127	0.48±0.276	0.30±0.317	0.43±0.275	0.41±0.293	0.35±0.303	0.60±0.085
848	0.05±0.189	0.20±0.282	0.09±0.221	0.16±0.299	0.22±0.305	0.44±0.275	0.49±0.215
873	0.08±0.195	0.45±0.292	0.11±0.230	0.19±0.320	0.40±0.278	0.39±0.318	0.54±0.184
880	0.00±0.000	0.64±0.051	0.49±0.269	0.37±0.292	0.40±0.196	0.54±0.287	0.58±0.186
Overall	0.03±0.130	0.33±0.296	0.22±0.284	0.26±0.289	0.37±0.275	0.36±0.294	0.55±0.153

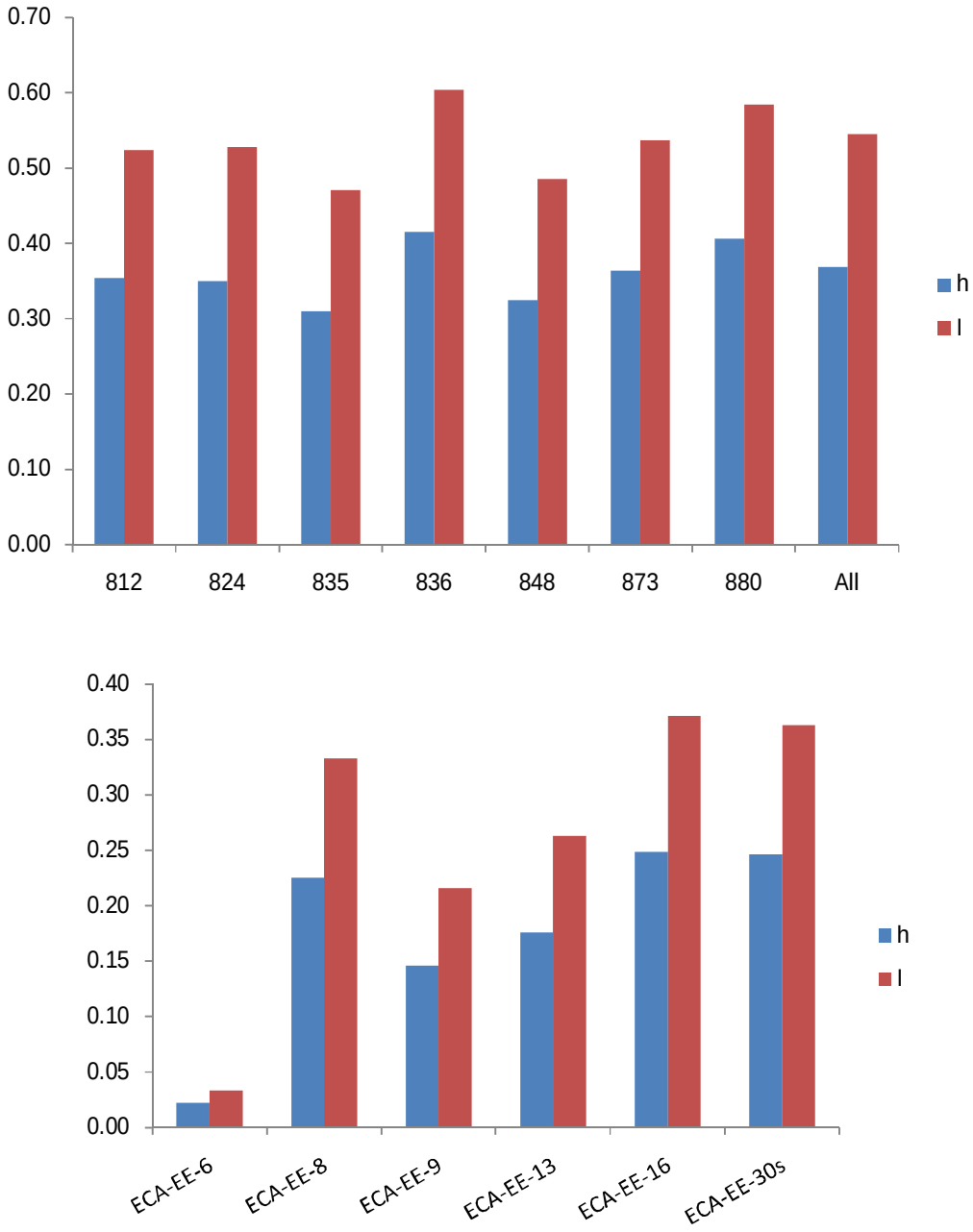


Figure 12. Nei's gene diversity (h) and Shannon's information index (I) of the selected primers (top) and the identified populations based on pedigree infomation (bottom).

5.8.3. Cluster analysis

According to Jaccard's similarity coefficient, 88% (523 out of 595) of the pairwise comparisons showed 25%–60% similarities (Figure 13). Specifically, about a quarter (142 out of 595) of the pairs had 45%–50% similarity. The maximum similarity was observed between L02 and L03 (96%) followed by L03 and L04 (94%) which were grouped in the same population named ECA-EE-6. The smallest similarity was recorded between L05 and L19 (27%) which were the members of population ECA-EE-8 and ECA-EE-9 respectively.

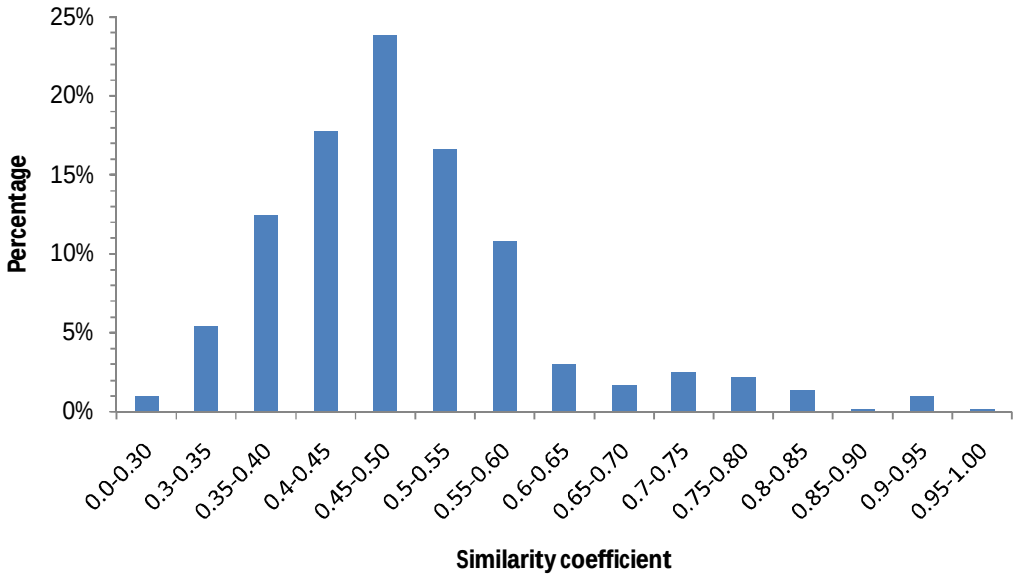


Figure 13. Percentage of Jaccard's similarity coefficient of 35 maize inbred lines genotyped by seven ISSR primers

The UPGMA dendrogram of the 35 inbred lines revealed seven populations at approximately 54% phenon level which was almost identical to the grouping based on pedigree information (Figure 14). All the individuals in each of the identified population according to their pedigree were clustered together in the dendrogram constructed using UPGMA analysis except five lines viz. L07 of ECA-EE-8, L18 of ECA-EE-9, L28 of ECA-EE-13, L30 and

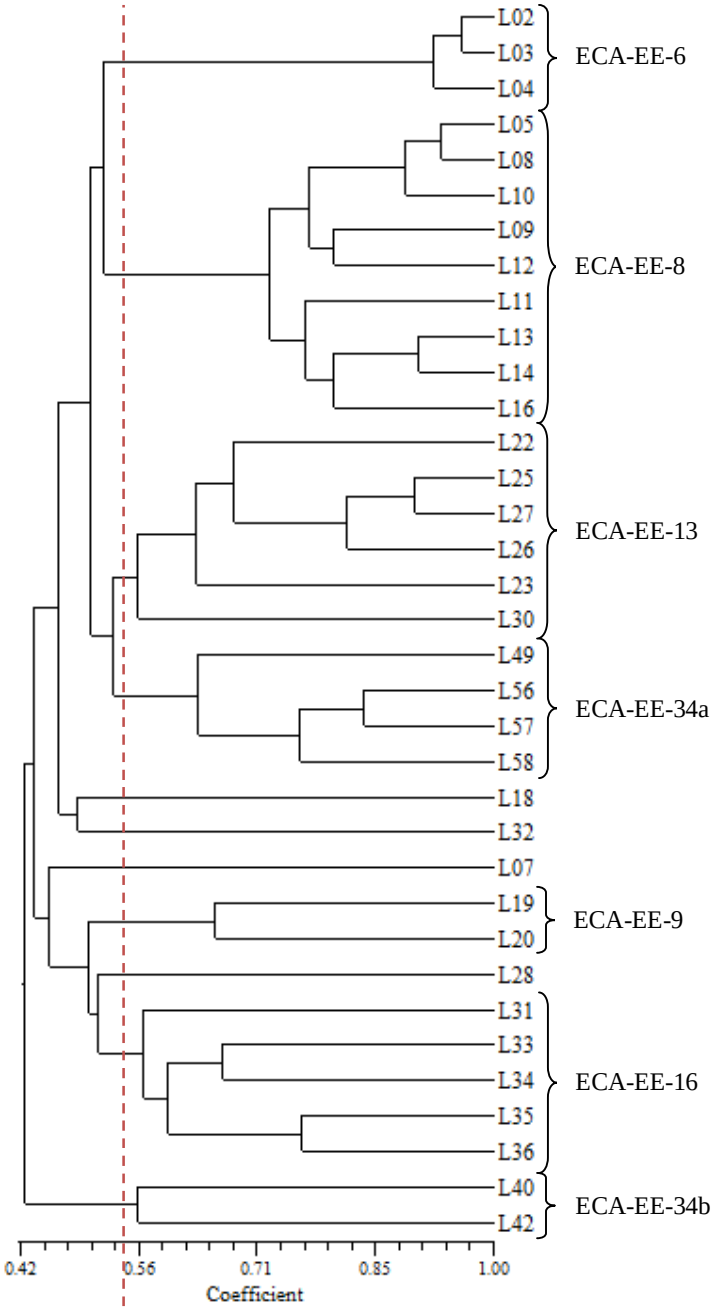


Figure 14. UPGMA tree for 35 QPM inbred lines using seven ISSR primers

L32 of ECA-EE-16. Among these, four lines couldn't be clustered to any of the populations as they stood out alone while L30 which was derived from ECA-EE-16 male parent was clustered with a population containing inbred lines derived from ECA-EE-13 male parent. Inbred lines L40 and L42 were grouped together isolated from the other populations to form

another group which is named ECA-EE-34b showing only 42% of genetic similarity with the others.

5.8.4. Principal coordinates analysis (PCoA)

Principal coordinates analysis indicated that the first two principal coordinates accounted for 21.3% of the total variation. The clustering of inbred lines using this analysis (Figure 15) was more or less in agreement with that of UPGMA analysis. The populations ECA-EE-6 and ECA-EE-8 which were separated into distinct groups using UPGMA analysis were also clearly separated from the rest of the inbred lines in case of PCoA forming two distinct groups and consisted of three and nine inbred lines respectively as the UPGMA analysis. The other inbred lines fell into another one distinct population which had separated into four populations in case of UPGMA analysis.

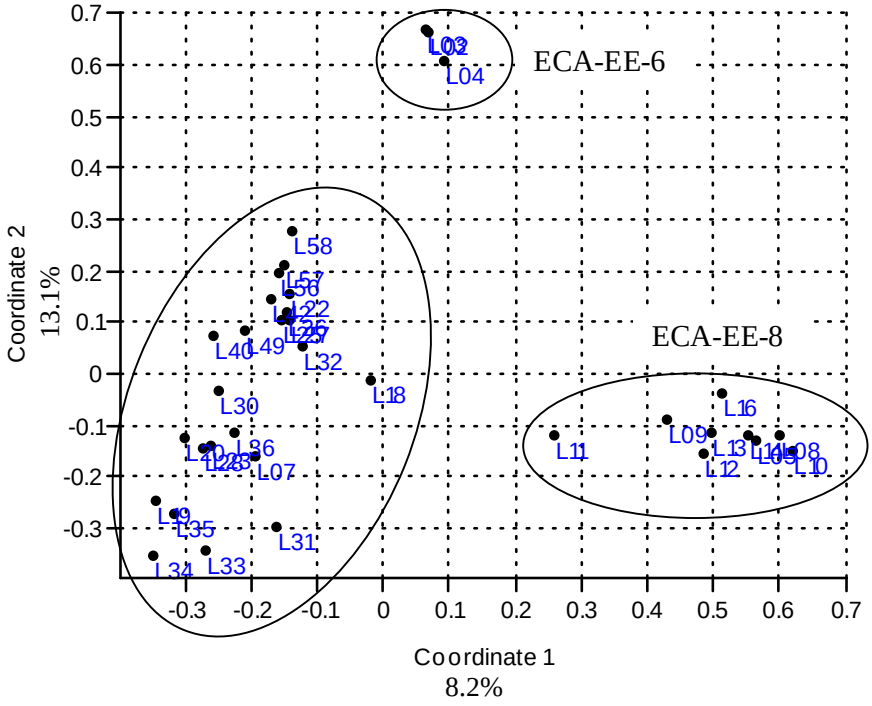


Figure 15. Principal coordinates analysis of 35 maize inbred lines based on seven ISSR primers

6. Discussion

6.1. Inbred line and hybrid performance

The foundation of any effective breeding program is the presence of genetic variability from which selection progress can be achieved for higher grain yield and improved agronomic traits. Significant differences observed among the inbred lines and hybrids for grain yield and most considered agronomic traits at each and across locations proved that genetic variations existed among the genotypes to allow good progress from selection for improvements of those traits. Several researchers observed such variations among genotypes as well (Legesse Welde *et al.*, 2009; Musila *et al.*, 2010; Badu-Apraku *et al.*, 2013; Aminu *et al.*, 2014; Rovaris *et al.*, 2014; Badu-Apraku *et al.*, 2015; Gudeta Nepir *et al.*, 2015). Non-significant differences were observed for RL among hybrids and inbred lines while significant for SL. Bhatnagar *et al.* (2004) and Musila *et al.* (2010) also obtained similar results which might show that shoot lodging was more important than root lodging.

Considerably high percentage of stalk lodging was recorded at Edo Gojola (up to 78.71% for the hybrids and up to 62.46% for the inbred lines) and Mieso (up to 56.38% for the hybrids and up to 68.12% for the inbred lines) than at MARC showing the potential of the sites to screen for SL. The significant differences observed among sites for all traits except CLR show that the testing sites had unique edaphoclimatic characteristics (Badu-Apraku et al., 2013; Rovaris et al., 2014; Badu-Apraku et al., 2015). The significant genotype × site and hybrid × site interactions for GY, SL, EL, KPR, CLR and PA imply that the hybrids did not perform consistently across such unique sites as also indicated by Dagne Wegary (2008) and Berhanu Tadesse (2009). For the other traits, the observed non-significant interaction effect indicates stable performance of the hybrids for those particular traits across the testing sites which is desirable to identify superior genotypes for wider adaptation.

The significant difference among checks for only EH, EPO, TKW, DA, CLR and PA indicates that the checks were variable only for these traits. The fact that the contrast between hybrids and checks was significant only for TKW and CLR suggests that the hybrids and checks were similar in performance for most of the traits. The variabilities among the checks and the similarities between hybrids and checks were stable across the testing sites as indicated by the non-significant check × site and hybrid vs check × site interactions. Moreover, the significant difference among the lines for all considered traits shows that there was genetic variability among the lines and the lines had differential responses under different sites for GY, PH, SL, DA, EH, EPP, EA and PA as observed from the significant line × site interactions for these traits. The significant differences among the testers only for GY, PH and TKW suggests that the testers, CML144 and CML159, had similar per se performances for most of the measured traits except these three. The significant contrast between lines and testers was observed only for EL, KPR, DA and EPP suggesting the similarity between lines and testers in per se performance for most of the traits. The lack of significant testers by site and lines versus testers contrast by site interactions for all measured traits suggests that performance of the testers and differences between lines and testers were stable and was not affected by site specific variations.

6.2. Combining ability and heterosis

The combining ability analysis showed significant line GCA while non-significant line × tester SCA for GY, DA, PH, EH, PA, EL and TKW indicating that the variability observed among the hybrids was attribuable to additive effects but not non-additive gene effects for most traits (Musila *et al.*, 2010; Shushay Welderufael *et al.*, 2013; Amare Seyoum *et al.*, 2016). If SCA mean square is not significant, Baker (1978) pointed out that the performance of a single cross progeny can be adequately predicted on the basis of GCA and so two parents

having the highest GCAs can be crossed to produce the best performing progenies. This was illustrated in this study when the lines with significantly positive GCA effects for GY like L52 (0.54), L35 (0.83), L45 (0.68) and L53 (0.63) were crossed with the tester with significantly positive GCA value for GY (CML159, 0.27) produced high yielding single crosses viz. L52/CML159 (5.38 t ha^{-1}), L45/CML159 (4.88 t ha^{-1}), L35/CML159 (5.02 t ha^{-1}) and L53/CML159 (4.85 t ha^{-1}). Non-significant SCA observed in this study is possibly a result of using parents that are related (Hill Jr., 1983; Musila *et al.*, 2010). However, both GCA and SCA were significant for EPO, SL, EA, KPR, ASI, CLR and RPE indicating that both additive and non-additive gene actions were important in the inheritance of these traits. Other authors also observed similar results for different traits (Legesse Welde *et al.*, 2009; Aminu *et al.*, 2014; Gudeta Nepir *et al.*, 2015; Akula *et al.*, 2016). In this case, Baker (1978) suggests to assess the relative importance of GCA and SCA. The assessment in this study revealed the preponderance of additive gene action (GCA) for all these traits which is in agreement with many other authors (Vasal *et al.*, 1993a; Vasal *et al.*, 1993b; Legesse Welde *et al.*, 2009; Badu-Apraku *et al.*, 2013; Aminu *et al.*, 2014; Gudeta Nepir *et al.*, 2015; Amare Seyoum *et al.*, 2016). Predominance of GCA over SCA effects suggests that hybridization, backcrossing and recurrent selection methods can be employed effectively on these inbred lines to develop hybrids, synthetics and populations. It also indicates that early generation testing may be effective and promising hybrids can be identified and selected based solely on the prediction from GCA effects (Badu-Apraku *et al.*, 2015)

The non-significant GCA of lines × site and testers × site interaction for grain yield and most other measured traits indicates the stability of combining abilities of the lines and testers under different sites. This result is partially in agreement with Gudeta Nepir *et al.* (2015) who found significant GCA of lines × site interaction but non-significant for GCA of testers × site interaction for most considered traits including GY. However, many authors found significant

line GCA × site interactions in contrast to the present study based on diallel crosses performance (Bhatnagar *et al.*, 2004; Pswarayi and Vivek, 2007; Musila *et al.*, 2010; Makumbi *et al.*, 2011; Badu-Apraku *et al.*, 2015). This might be due to genotype and site differences used in different studies. In the present study, the non-significant interaction of SCA with site for grain yield and most other measured traits indicates that performance of the hybrids were consistent in the varying testing sites. Previous authors also obtained similar results (Legesse Welde *et al.*, 2009; Musila *et al.*, 2010; Gudeta Nepir *et al.*, 2015). These results suggest that it is possible to select inbred lines and hybrids with stable performance across the diverse sites.

Superior inbred lines in terms of their estimate of GCA effect for grain yield and other agronomic traits are important for contribution of favorable alleles for that trait and the improvement of that trait in a population (Legesse Welde *et al.*, 2009; Makumbi *et al.*, 2011). Significant and positive GCA effects observed for GY of the inbred lines L35, L45, L53, L4, L21, L52 and L32 across sites indicates that these inbred lines can contribute favorable alleles for high GY to their progenies. In addition to GY, the significant and desirable GCA effects of inbred line L35 for ASI, PH, CLR, PA, EL and KPR; L45 for DA, EH, EPO, EA, KPR and RPE; L53 for DA, EL and TKW; L21 for ASI, PH and EL and L52 for DA, ASI, EL and KPR indicates that these inbred lines can also contribute favorable alleles for those traits and can be used to improve those traits in their progenies. Inbred lines L22, L39, L38, L27, L51, L58 and L12 can also contribute favorable alleles for earliness.

The positive SH of L52/CML159, L18/CML159 and L35/CML159 indicates that these hybrids are better than both checks and can therefore be released to end users and/or used for further breeding activities. Analysis of heterosis revealed that hybrids tend to be earlier than

the average of their parents and the better parent as demonstrated by negative better and mid parent heterosis for all of the hybrids.

Pswarayi and Vivek (2007) suggest maintenance of balance between earlier maturity and higher yield while selecting early maturing germplasm. In the current study, even though the earliest of all the hybrids was found to be the standard check, MH130, most of the hybrids were significantly earlier than the highest yielding standard check, MH140, which shows that the highest yielding hybrids were earlier than MH140. The hybrid L52/CML159 had higher yield than MH140 while significantly earlier.

This study showed also that the effect of heterosis was towards taller plants and higher ear height as shown by the significantly positive BPH for PH and EH. No hybrid was taller than MH140 and no hybrid had lower EH than MH130 indicating that these two hybrid checks had the extreme values for PH (MH140) and EH (MH130) which can be due to the lateness of MH140 and earliness of MH130 as demonstrated by the significantly positive correlation among DA, PH and EH.

The tester CML159 was better than CML144 as a parent to stimulate heterosis in its hybrids for GY and most of the measured traits while CML144 was better to induce heterosis for yield components.

6.3. Variability and traits association in QPM genotypes and their implications for selection

The existence of variability is essential for wide adaptability in the genotypes. Selection is effective when there is genetic variability among the individuals in a population (Vashistha *et al.*, 2013). Variability estimates in the present study indicated that PH, EH, SL, EL and TKW showed the highest phenotypic and genotypic variances. Previous authors also found similar

results for these traits (Yusuf, 2010; Nzuve *et al.*, 2014; Sesay *et al.*, 2016). Environmental variance was higher than the other variance components for all studied traits except DA and ASI where phenotypic variance was the highest. It suggests that most of the characters were greatly influenced by the environments. Similar results were observed by Ojo *et al.* (2006) and Kashiani *et al.* (2010).

Direct selection for high GY under stress or non-stress conditions is not effective mostly as it has low to moderate heriatbility while indirect selection for high GY using secondary traits is effective strategy (Bänziger *et al.*, 2000). Moreover, heritability values when coupled with estimates of GA are more useful as a selection tool for high GY (Johnson *et al.*, 1955). Therefore, study of heritability, GA and correlation is used as one of the tools to determine the value of secondary traits in relation to GY (Hallauer *et al.*, 2010) and to select best performing hybrids.

In this study, GY had highly significant negative correlation to SL. This is consistent with the results of Musila *et al.* (2010) across well watered environments. Negative correlation was also observed between GY and DA which is also in agreement with the results of Musila *et al.* (2010) under well watered condition. This can be due to the indirect influence of DA on SL as exhibited by the highly significant positive correlation between DA and EH and the highly significant positive correlation between EH and SL. In other words, late maturing hybrids had high ear placement which contributed to high SL. Therefore, highly significant negative correlation between SL and GY may have caused the negative correlation between DA and GY.

On the other hand, when the observed significant positive correlation between EH and SL was compared to the low correlation of PH with SL, it may show that the determinant factor for SL was EH rather than PH. Hence, it can be possible to select tall hybrids with low ear

placement and therefore with low shoot lodging percentage and high yield. This can be clearly demonstrated by the fact that the top yielding hybrids had low ear placement which can illustrate the balance between earlier maturity and higher yield that Pswarayi and Vivek (2007) suggested. This balance is extremely important for farmers to use the long stalk for fencing, house construction, feed and other purposes without yield penalty due to lodging in windy areas and seasons.

The highest positive correlation was observed between GY and TKW followed by EL and PH while strong negative associations were noticed between GY and EA, DA, ASI, EPO and SL. Among these traits, TKW and EPO had moderately high heritability and moderate GA while DA and SL had moderately high heritability and high GA. EA exhibited medium heritability and moderate GA. This result is in agreement with the findings of Sesay *et al.* (2016) and implies that effective indirect selction can be performed using these secondary traits. It is also an indicator of the preponderance of additive gene action in these characters (Sesay *et al.*, 2016).

6.4. Molecular diversity among QPM inbred lines

6.4.1. SNP markers and their informativeness

As SSRs are multi-allelic markers, they can provide many times the number of alleles per locus. However, SNPs are less informative compared to SSRs because they are biallelic. However, if enough SNPs are used the difference in the informativeness of the markers would be overcome (Hamblin *et al.*, 2007). Laval *et al.* (2002) concluded that ($k - 1$) times more SNP markers are needed to achieve the same genetic distance accuracy as a set of SSRs with k alleles. Another author also proposed that between 7–11 times more SNPs than SSRs should be used for analyzing population structure and genetic diversity (Van Inghelandt *et*

al., 2010). Therefore, the 165 SNPs used in this study can be assumed to be similar to a study using about 15–20 SSRs with an average of two SSRs per chromosome. This number of markers is enough for the current diversity analysis as the studied materials are inbred lines and small in number. Moreover, the SNPs used in this study are identified from three studies taking only those that are common in their recommendations (Lu *et al.*, 2009; Kassa Semagn *et al.*, 2012; Dao *et al.*, 2014). These authors used 1537 SNPs and recommended a subset that are more informative to be used in routine genetic diversity studies in maize based on the quality of the SNPs.

The average PIC value in the present study (0.298) was similar to the highest reported value of 0.31 by Yang *et al.* (2010) using 926 informative SNPs in 527 inbred lines with diverse backgrounds and was higher than a value of 0.239 reported by Hao *et al.* (2011) using 1006 polymorphic SNPs in 80 maize lines in China, a value of 0.259 reported by Lu *et al.* (2009) using 1034 informative SNPs in 770 diverse maize inbred lines and a value of 0.256 reported by Dao *et al.* (2014) using 1057 informative SNPs in 100 diverse maize inbred lines. Similarly, the mean gene diversity value in this study (0.376) was consistent with the value of 0.39 reported by Yang *et al.* (2010) but higher than the values 0.290 and 0.321 reported by Hao *et al.* (2011) and Lu *et al.* (2009), respectively. Since PIC value refers to the polymorphic level of each SNP with respect to the amount of polymorphisms exhibited and gene diversity or expected heterozygosity refers to the probability that two randomly chosen alleles from the population are different, the higher PIC and gene diversity values in this study can be attributed to the utilization of selected highly polymorphic SNPs in this study from different studies exploring maize inbred lines.

According to Patterson *et al.* (2006) the central challenges in analyzing genetic dataset are knowing whether a given population is homogeneous or composed of distinct subgroups and

finding quantifiable evidence for these subgroups from the data. In order to answer these detection and quantification questions, the model based population structure analysis, NJ-cluster analysis and principal component analysis were used to explore the extent of genetic differentiation, population structure and patterns of relationship among 58 QPM inbred lines. All these different multivariate methods revealed that there were seven populations which were almost identical to the identified populations based on *a priori* pedigree knowledge. Most of the lines with the same female parents, as all the lines shared the same male (recurrent) parent, were clustered into the same group indicating the powerfulness of keeping pedigree information to identify inbred lines. This result was in agreement with previous authors (Lu *et al.*, 2009; Wen *et al*., 2011; Kassa Semagn *et al.*, 2012; Dao *et al.*, 2014).

Even though earlier works concluded that there was high concordance between PCA and model-based population structure and low concordance between cluster analysis and the other multivariate analyses (Kassa Semagn *et al.*, 2012; Dao *et al.*, 2014), the present study demonstrated high consistency in all of the analyses. All of them clearly identified the same seven distinct populations and revealed similar membership of inbred lines in each population. This may be due to the small number of inbred lines represented in each population in this study that can reduce the number of different combinations of genetic distance which otherwise would produce several alternative clusters and thereby ambiguity to select the best one which was described by the other authors as the reason for the inconsistency shown by cluster analysis.

In the identified populations, the CML testers CML144 and CML159 were identified as mixed and stood out together as revealed by having a common branch in the dendrogram and being placed near each other in the PC plot. However, these inbred lines were described as belonging to different heterotic groups based on diallel or line × tester analysis by CIMMYT

breeders. CML144 belonged to heterotic group B and CML159 to heterotic group A (Machida *et al.*, 2010). The closeness of the testers in molecular diversity analysis even though belonging to different heterotic groups based on heterosis was in agreement with previous authors who concluded that no clear population structure was revealed for most studied inbred lines in heterotic groups A and B because of the mixed genetic constitution of the original germplasm (Lu *et al.*, 2009; Wen *et al.*, 2011; Kassa Semagn *et al.*, 2012; Dao *et al.*, 2014). Moreover, it was not possible to decide the heterotic patterns of the inbred lines in this study based on the combining ability analysis result because of the non-significant SCA for grain yield and most of the traits considered.

There were 3.7% (64 out of 1711) of pairwise comparisons that had genetic distance less than 0.05 which indicates that there were small number of redundant lines while most of the distance fell between 0.35 and 0.5 which was greater than that of Kassa Semagn *et al.* (2012) who reported the majority fell between 0.3 and 0.4. Therefore, this result suggests that each line in this study is potentially contributing new alleles to a breeding program (Kassa Semagn *et al.*, 2012; Dao *et al.*, 2014).

6.4.2. ISSR markers and their informativeness

The success in generating wide range of polymorphic loci depends on proper choice of primers for DNA amplification and the optimum number of primers. Few primers are enough to generate useful information when variation in genotypes is high (Lenka *et al.*, 2015). In view of the high variation of inbred lines in this study and high level of polymorphism (97.73%), the number of ISSR markers used in this study (7 selected out of 24 examined) can be considered enough to distinguish among the lines. Idris *et al.* (2012) used the same number of ISSR primers to successfully assess genetic diversity among nine maize genotypes even though they found lesser level of polymorphism (69%). However, high level of

polymorphism was reported also by Kantety *et al.* (1995) (95%) using 10 ISSR primers to detect polymorphism among 19 popcorn and 8 dent maize inbred lines and by Lenka *et al.* (2015) (94.87%) using 12 ISSR primers to amplify DNA of 49 inbred lines of maize.

The number of scorable bands reported in the present study ranged from 10 to 18 bands with a mean of 12.6 bands per primer. Carvalho *et al.* (2002) reported 5 to 17 fragments with 9.5 fragments per primer; Idris *et al.* (2012) found 4 to12 bands with 6.5 bands per primer; Lenka *et al.* (2015) recorded 4-9 bands with 6.5 bands per primer and do Amaral Júnior *et al.* (2011) discovered 4 to 11 bands with 8.1 bands per primer. The differences in all of these studies can be attributable to the differences in type and number of primers, primer length, GC content, melting temperature and annealing temperature used in each study which are considered as critical factors for DNA amplification (Lenka *et al.*, 2015).

The ISSR primer 812 ($(GA)_8A$) had the highest number of bands with 94.44% of polymorphism. Idris *et al.* (2012) and do Amaral Júnior *et al.* (2011) found similar results by ISSR primer 810 ($(GA)_8T$) with 12 and 11 total number of bands respectively. Both studies reported percentage of polymorphic bands of 92%. Carvalho *et al.* (2002) also found that primers based on $(GA)_9$ produced the greatest number of bands. This indicates that the bi-nucleotide microsatellite $(GA)_n$ is a frequent repeat well distributed in maize genome since the potential of ISSR markers to generate polymorphic bands depends on the microsatellite frequency and distribution in the genome of a species (Morgante and Olivieri, 1993). Sequence studies in different crops revealed that regions containing (GA) repeats were frequent in the genome of different crops. Taramino and Tingey (1996) revealed that seven out of 34 sequences analyzed had regions containing $(GA)_n$ in maize. This sequence was also frequent in *Vigna* (Ajibade *et al.*, 2000), and in diploid, tetraploid and hexaploid wheat (Nagaoka and Ogihara, 1997).

Molecular markers allow direct comparison of similarity of genotypes at DNA level. Genetic similarity between two genotypes, populations or individuals may be calculated by various statistical measures depending on the data set (Mohammadi and Prasanna, 2003). Jaccard's coefficient of similarity was preferred in this study because it gives more weight to matches than mismatches (Sneath and Sokal, 1973); it is less biased by the occurrence of a given level of artificial bands (Lamboy, 1994) and sharing or matching of a band has a direct biological meaning in that it is an estimate of the expected properties of amplification. The similarity coefficient in this study ranged from 0.265 to 0.961 with an average of 0.496 which indicates wide genetic diversity among the inbred lines.

When the dendrogram constructed by UPGMA analysis using Jaccard's similarity matrix from seven ISSR primers is compared to the dendrogram constructed by NJ analysis using Roger's genetic distance from 165 SNPs, membership of 71.4% of the inbred lines in the identified populations in each dendrograms was similar. Similarly, the dendrogram constructed from the ISSR primers had 85.7% of the inbred lines clustered in the same populations as the *a priori* predefined populations based on pedigree information.

The least variability was observed within the population derived from ECA-EE-6 as can be clearly observed from its 5.68% polymorphism, Nei's gene diversity value of 0.03 and Shannon's information index of 0.02. It had only three member inbred lines. These inbred lines viz. L02, L03 and L04 can be considered as redundant lines as indicated by the 96% similarity between L02 and L03, 94% similarity between L03 and L04 and 91% similarity between L02 and L04. The highest genetic variability was observed in the population derived from ECA-EE-16.

7. Conclusion

In the present study, inbred lines L35, L45, L53 and L52 had desirable significant GCA effects for grain yield and other agronomic traits and could be used in the breeding program for the development of high yielding hybrids with desirable agronomic performance. Additive gene action was more important than non-additive gene action for the measured traits, indicating that GCA was the major component accounting for the differences among the inbreds evaluated in the present study.

Moreover, hybrid combination L52/CML159 had the best SCA effects for grain yield and other most important traits and the maximum standard heterosis over MH140. The parents of this hybrid variety – L52 and CML159 (tester) had the best *per se* performances and GCA effects. Therefore, this single cross hybrid was identified as the most stable and high yielding across environments.

Therefore, this study identified good combining QPM inbred lines and a good performing QPM hybrid combination for future breeding activities in drought stressed areas of Ethiopia and direct release for the target environment. In general, the information from this study will be useful for researchers who intend to develop varieties with high grain yield and desirable agronomic characters.

The present study showed that root lodging was not important characteristics to be considered while shoot lodging played an important role in determining grain yield. It also showed that ear height is more important than plant height to develop high yielding hybrids and that it is possible to select high yielding varieties which are early but tall with low ear placement. Moreover, secondary traits like thousand kernl weight, ear position, days to anthesis and

shoot lodging had strong correlation with grain yield with moderately high heritability and moderate to high genetic advance upon selection.

The SNP markers used in this study were very effective to characterize the inbred lines. Multivariate analyses using these SNPs showed clear separation of the inbred lines into seven clusters which are in agreement with pedigree information. Hence, pedigree information was proved to be a powerfull tool to cluster related inbred lines. This study also alluded to the fact that it was not possible to cluster inbred lines into CIMMYT identified heterotic groups based on both phenotypic (SCA) and SNP data. There were high genetic distance coefficients among most pairs of lines in this study indicating clearly that the majority of the inbred lines in this study were unique.

Moreover, the results in the present study demonstrate the efficiency of ISSR molecular markers in the separation and identification of the variability of maize inbred lines evaluated in the study. ISSR markers were found to be as effective as SNP markers in clustering inbred lines into sub-groups which are in agreement with pedigree information. The ISSR result in this study revealed that primers based on $(GA)_n$ were highly polymorphic as they are abundent in maize genome.

In general, the results from this diversity study based on SNP and ISSR markers will be useful to breeders in selecting best parental combinations for starting new breeding populations, mapping population and marker assisted breeding.

8. Recommendations

The favorable alleles from the selected inbred lines based on their GCA should be introgressed into QPM breeding populations developed for drought stressed areas for improvement of the target traits. The F_1s produced for this study could further be used to develop F_2 and advanced populations for development of better inbred lines for future hybrid development, especially single cross hybrids with vigorous parents for release. In addition, the identified best single cross hybrid – L52/CML159 should be extensively evaluated in on-station and on-farm trials with other varieties to confirm the consistency of its performance and release. Moreover, it can be used for further breeding.

When the very important role that shoot lodging plays in the productivity of the genotypes in this study is considered, it should be included as a major criteria in selecting high yielding varieties. Emphasis also should be given to reduced ear height rather than reduced plant height in order for farmers to have high yielding early-yet-tall varieties. Simultaneously, the high importance of days to anthesis, plant and ear height coupled with the result found in this study that MH140 was the tallest hybrid while MH130 was the earliest with low ear placement strongly advise breeders for the drought stressed areas to focus in the development of taller yet early hybrids with lower ear placements than the check varieties in order to continuously improve these traits and cater for the needs of farmers in their target environments.

Further study should be conducted to classify these inbred lines into heterotic groups for future breeding plan. Efforts should also be made to identify divergent heterotic groups of available inbred lines in the breeding project for drought stressed areas of Ethiopia as the already used heterotic pattern was not convenient to cluster inbred lines both genotypically and phenotypically. Yet, heterotic patterns are useful tools for increasing the efficiency of

breeding programs through organizing the large number of inbreds being developed by the breeding project.

The SNPs used in this study for low to medium density genotyping in maize inbred lines are recommended to be used in uniplex assays like KASP^{TM}. In addition, ISSR markers are recommended for quick and preliminary characterization of maize inbred lines to evaluate their diversity.

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10. Appendices

Appendix 1. Designation, pedigree and tryptophan content of the inbred lines

Label	Pedigree	Pop	Trp w/w (%)*
L1	(ECA-EE-6/PL15QPMC7SRC1F2//POOL15QPMSR)-B-14-#-1-2-2-B-1-#	A	0.084
L2	(ECA-EE-6/PL15QPMC7SRC1F2//POOL15QPMSR)-B-45-#-2-1-1-B-2-#	A	0.100
L3	(ECA-EE-6/PL15QPMC7SRC1F2//POOL15QPMSR)-B-45-#-2-1-3-B-1-#	A	0.102
L4	(ECA-EE-6/PL15QPMC7SRC1F2//POOL15QPMSR)-B-45-#-2-1-4-B-1-#	A	0.098
L5	ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR-B-28-#-1-1-1-B-3-#	B	0.087
L6	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-1-2-B-1-#	B	0.087
L7	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-2-1-B-2-#	B	0.090
L8	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-2-1-B-3-#	B	0.092
L9	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-2-2-B-2-#	B	0.081
L10	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-2-2-B-3-#	B	0.092
L11	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-2-3-B-2-#	B	0.087
L12	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-1-2-4-B-1-#	B	0.091
L13	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-2-3-1-B-1-#	B	0.083
L14	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-3-4-1-B-2-#	B	0.073
L15	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-3-4-2-B-2-#	B	0.077
L16	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-28-#-3-4-3-B-2-#	B	0.083
L17	(ECA-EE-8/PL15QPMC7SRC1F2//POOL15QPMSR)-B-53-#-1-2-3-B-2-#	B	0.106
L18	(ECA-EE-9/PL15QPMC7SRC1F2//POOL15QPMSR)-B-71-#-1-3-1-B-2-B	C	0.089
L19	(ECA-EE-9/PL15QPMC7SRC1F2//POOL15QPMSR)-B-71-#-1-3-1-B-3-B	C	0.088
L20	(ECA-EE-9/PL15QPMC7SRC1F2//POOL15QPMSR)-B-71-#-1-3-2-B-1-B	C	0.086
L21	(ECA-EE-9/PL15QPMC7SRC1F2//POOL15QPMSR)-B-71-#-1-3-2-B-2-B	C	0.097
L22	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-6-#-2-1-2-B-1-#	D	0.090
L23	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-1-1-B-1-B	D	0.106
L24	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-1-2-B-2-#	D	0.107
L25	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-1-3-B-1-#	D	0.116

Appendix 1. Inbred lines continued

Label	Pedigree	Pop	Trp w/w (%)*
L26	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-1-4-B-2-#	D	0.121
L27	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-2-1-B-3-#	D	0.120
L28	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-2-3-B-3-#	D	0.121
L29	(ECA-EE-13/PL15QPMC7SRC1F2//POOL15QPMSR)-B-24-#-1-2-4-B-2-#	D	0.125
L30	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-1-2-1-B-1-#	E	0.099
L31	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-1-4-1-B-1-#	E	0.084
L32	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-1-5-2-B-3-#	E	0.094
L33	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-1-5-3-B-3-#	E	0.109
L34	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-2-1-2-B-2-#	E	0.091
L35	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-2-2-1-B-1-#	E	0.092
L36	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-2-2-2-B-1-#	E	0.099
L37	(ECA-EE-16/PL15QPMC7SRC1F2//POOL15QPMSR)-B-85-#-2-2-3-B-1-#	E	0.089
L38	(ECA-EE-31/PL15QPMC7SRC1F2//POOL15QPMSR)-B-58-#-2-1-1-B-1-#	F	0.103
L39	(ECA-EE-33/PL15QPMC7SRC1F2//POOL15QPMSR)-B-82-#-1-2-2-B-1-#	F	0.087
L40	(ECA-EE-33/PL15QPMC7SRC1F2//POOL15QPMSR)-B-82-#-1-2-3-B-1-#	F	0.093
L41	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-10-#-1-2-1-B-1-#	G	0.107
L42	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-1-3-2-B-1-#	G	0.109
L43	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-1-3-2-B-3-#	G	0.101
L44	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-1-3-3-B-1-#	G	0.105
L45	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-1-3-3-B-2-#	G	0.096
L46	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-3-5-1-B-1-#	G	0.095
L47	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-3-5-2-B-1-#	G	0.099
L48	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-20-#-3-5-3-B-2-#	G	0.092
L49	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-2-1-B-1-#	G	0.092
L50	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-2-2-B-1-#	G	0.088
L51	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-2-3-B-2-#	G	0.121
L52	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-4-1-B-1-#	G	0.112

Appendix 1. Inbred lines continued

Label	Pedigree	Pop	Trp w/w (%)*
L53	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-4-1-B-2-#	G	0.088
L54	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-4-2-B-2-#	G	0.089
L55	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-4-3-B-2-#	G	0.120
L56	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-5-1-B-1-#	G	0.085
L57	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-5-1-B-2-#	G	0.093
L58	(ECA-EE-34/PL15QPMC7SRC1F2//POOL15QPMSR)-B-86-#-1-5-2-B-2-#	G	0.084
T1	P62-C5-FS182-2-1-2-B-B-3-1-B (CML144)	H	-
T2	P63-C2-FS5-1-3-1-B-2-1-1-B (CML159)	H	-

**The values are expressed in w/w = grams per 100 grams of sample.*

Appendix 2. Combined data (Sorted by RelGY) for grain yield and other related traits in quality protein maize hybrids evaluated at Melkassa, Edo Gojola and Mieso during 2014 main season

Entry	Pedigree	Across			Across	Anth	ASI	Plant	Ear	Ear	Lodging		Ears/	P.sorg	Ear	Plant	ED	EL	KPR	RPE	TKW
		RelGY*	Rank		Grain Yield	Date		Height	Height	Position	Root	Stem	Plant		Aspect	Aspect					
		%	Avg*	StdDev*	t/ha	d	d	cm	cm	0-1	%	%	#	1-5	1-5	1-5					
110	L52/CML159	131	12	18	5.38	70.7	-0.4	229.5	94.6	0.44	0.1	0.9	0.97	2.4	2.4	2.5	42.02	162.31	35.59	14.77	234.47
47	L18/CML159	124	18	21	5.07	71.8	1.3	239.7	108.9	0.49	3.1	4.3	0.99	2.5	2.1	2.2	44.17	154.64	34.21	14.38	206.18
93	L35/CML159	123	11	11	5.02	72.4	0.3	233.7	110.9	0.50	0.2	4.2	1.04	2.0	2.8	2.1	41.64	164.23	34.49	14.27	240.34
95	L37/CML159	119	13	8	4.89	73.0	0.1	245.6	113.4	0.50	0.0	2.1	1.09	2.4	2.3	2.3	42.08	157.85	33.54	13.90	226.38
103	L45/CML159	119	17	13	4.88	71.3	1.7	228.2	83.3	0.40	1.4	4.6	0.97	2.4	2.2	2.4	44.12	150.99	35.95	15.27	207.38
115	L57/CML159	119	15	7	4.86	71.3	0.3	217.8	103.1	0.51	3.2	2.3	0.88	2.4	2.3	2.4	41.84	153.89	34.11	13.80	246.10
111	L53/CML159	119	23	19	4.85	70.7	2.0	233.6	104.3	0.49	0.1	9.7	0.87	2.5	2.5	2.5	41.90	152.84	30.71	14.13	236.10
92	L34/CML159	118	33	49	4.83	72.0	-0.2	235.9	108.0	0.49	-0.1	0.7	0.85	2.5	2.7	2.5	41.16	166.02	35.44	13.20	241.14
4	L4/CML144	115	32	43	4.67	74.3	-0.3	234.2	110.4	0.55	1.5	6.0	0.92	2.4	2.6	2.2	43.81	155.42	35.35	14.33	204.80
116	L58/CML159	114	28	32	4.66	71.1	-0.2	220.2	99.7	0.48	0.0	0.8	0.92	2.5	2.2	2.5	38.99	146.58	33.20	13.39	238.38
61	L32/CML144	114	26	16	4.66	73.4	-1.1	236.3	111.0	0.50	3.5	3.8	1.50	2.4	2.1	2.6	42.29	155.40	35.33	14.51	206.92
64	L35/CML144	113	34	27	4.62	73.5	-1.0	240.9	116.0	0.51	1.8	5.0	1.18	2.5	2.4	2.1	41.00	163.55	36.04	14.46	191.71
34	L5/CML159	113	25	12	4.62	72.3	2.2	226.9	99.2	0.47	-0.1	3.3	0.88	2.5	2.3	2.3	44.62	147.95	32.66	14.20	240.13
50	L21/CML159	112	28	15	4.60	71.6	0.6	243.6	107.6	0.47	6.7	12.7	1.03	2.5	2.7	2.3	43.75	164.82	34.58	14.91	211.76
97	L39/CML159	112	28	22	4.59	69.8	0.5	229.5	102.3	0.48	-0.2	4.2	0.95	2.4	2.6	2.6	41.74	152.46	35.72	13.93	239.47
89	L31/CML159	112	29	20	4.57	70.9	1.0	229.7	98.3	0.46	0.1	4.9	0.96	2.1	2.7	2.4	42.29	140.83	34.84	14.73	219.63
82	L53/CML144	112	34	34	4.57	71.7	0.7	225.5	118.0	0.56	1.5	7.8	1.20	2.5	2.7	2.4	39.77	160.99	35.72	14.47	200.04
96	L38/CML159	111	28	9	4.55	68.6	1.5	217.7	80.9	0.39	4.9	10.4	0.97	2.5	3.0	2.5	42.22	152.24	36.19	14.90	210.50
112	L54/CML159	111	29	14	4.55	70.5	-0.2	223.0	100.9	0.49	0.2	5.4	1.01	2.6	2.5	2.5	40.13	151.09	32.76	14.12	240.22
99	L41/CML159	111	31	15	4.55	71.9	2.9	233.4	114.1	0.52	5.0	7.4	1.02	2.4	2.2	2.1	46.56	147.80	32.96	15.26	222.94

Appendix 2. Combined data of QPM hybrids continued

Entry	Pedigree	Across			Across	Anth	ASI	Plant	Ear	Ear	Lodging		Ears/	P.sorg	Ear	Plant	ED	EL	KPR	RPE	TKW
		RelGY*	Rank		Grain Yield	Date		Height	Height	Position	Root	Stem	Plant		Aspect	Aspect					
		%	Avg*	StdDev*	t/ha	d	d	cm	cm	0-1	%	%	#	1-5	1-5	1-5					
2	L2/CML144	111	31	16	4.54	72.9	0.2	244.1	105.1	0.46	0.0	4.5	0.99	2.4	2.5	2.3	44.89	151.07	35.09	15.22	190.75
98	L40/CML159	111	35	33	4.53	71.3	-0.1	227.2	113.3	0.54	-0.1	1.4	1.09	2.3	2.6	2.6	40.48	151.69	35.48	13.31	229.04
49	L20/CML159	111	40	63	4.50	71.3	2.1	231.2	105.0	0.48	9.5	15.2	1.02	2.5	2.2	2.4	42.62	155.40	34.19	14.10	226.31
118	NA	110	40	58	4.50	73.8	1.7	225.3	115.0	0.53	1.5	3.1	0.99	1.6	2.6	2.1	43.96	155.44	32.49	14.49	238.67
108	L50/CML159	110	30	12	4.49	71.2	-0.2	216.7	97.0	0.48	0.0	4.8	1.02	2.5	2.7	2.4	41.78	157.63	34.58	14.00	230.14
3	L3/CML144	110	33	9	4.49	73.0	0.6	241.3	98.7	0.44	3.3	4.3	1.12	2.4	2.5	2.1	44.40	150.72	34.11	15.07	206.21
48	L19/CML159	109	42	31	4.44	73.3	2.0	243.0	106.8	0.47	11.8	8.5	0.99	2.5	2.8	2.5	42.86	168.07	34.20	14.90	219.47
119	NA	108	37	17	4.43	66.7	1.1	213.3	84.3	0.43	9.7	7.9	1.01	2.5	2.3	2.5	41.30	148.84	33.40	14.66	245.22
90	L32/CML159	108	34	6	4.43	70.9	0.3	227.5	105.5	0.49	6.4	6.6	1.13	2.3	2.5	2.5	42.85	150.56	34.27	13.92	225.41
106	L48/CML159	109	39	36	4.43	73.5	1.5	208.3	85.4	0.42	6.4	15.2	1.02	2.5	2.7	2.5	42.14	149.14	33.50	15.29	194.08
109	L51/CML159	108	35	20	4.42	71.1	0.7	223.8	101.3	0.49	1.5	2.1	1.12	2.1	2.4	2.5	40.36	151.77	34.82	13.99	228.72
88	L30/CML159	108	42	44	4.42	71.2	0.7	223.7	99.6	0.48	-0.1	1.8	0.99	2.5	2.3	2.3	44.19	145.83	32.81	14.35	217.03
33	L4/CML159	109	43	56	4.42	71.9	1.6	228.2	97.2	0.45	3.4	4.2	0.99	2.5	2.9	2.0	43.08	147.00	36.49	14.67	203.87
32	L3/CML159	108	37	8	4.41	71.4	2.0	242.6	92.4	0.42	0.0	0.5	0.96	2.6	2.9	2.1	42.27	151.34	33.83	14.31	215.55
66	L37/CML144	107	39	18	4.39	73.6	0.4	242.5	109.9	0.49	2.3	7.2	1.07	2.4	2.4	2.3	41.70	153.82	35.05	14.15	198.49
117	NA	107	41	25	4.37	74.5	0.8	224.5	95.7	0.45	1.6	11.6	1.17	2.3	2.9	2.5	42.81	148.74	33.78	14.60	206.89
57	L28/CML159	107	52	40	4.34	71.1	-0.1	233.9	114.1	0.52	3.3	16.3	0.96	2.5	3.0	2.5	43.27	156.91	34.75	13.97	214.89
43	L14/CML159	106	50	35	4.34	73.5	2.0	227.9	102.7	0.47	3.1	2.2	0.91	2.6	2.4	2.1	41.99	145.89	33.26	13.82	222.66
105	L47/CML159	106	47	25	4.33	72.7	2.6	224.3	96.9	0.46	1.5	10.6	1.04	2.5	2.5	2.6	41.61	145.83	34.37	14.40	219.84
104	L46/CML159	106	53	49	4.32	74.6	2.8	234.8	90.9	0.41	1.7	6.1	0.75	2.4	2.5	2.5	40.83	148.33	35.68	14.84	208.81
44	L15/CML159	105	45	24	4.32	73.9	3.6	230.4	101.6	0.48	3.2	0.3	0.99	2.4	2.6	2.1	43.81	141.76	33.30	14.94	201.08
60	L31/CML144	105	42	17	4.32	71.7	0.5	225.3	101.7	0.48	0.2	2.3	1.05	2.4	2.3	2.6	44.92	153.17	37.09	16.40	212.49

Appendix 2. Combined data of QPM hybrids continued

Entry	Pedigree	Across			Across	Anth	ASI	Plant	Ear	Ear	Lodging		Ears/	P.sorg	Ear	Plant	ED	EL	KPR	RPE	TKW
		RelGY*	Rank		Grain Yield	Date		Height	Height	Position	Root	Stem	Plant		Aspect	Aspect					
		%	Avg*	StdDev*	t/ha	d	d	cm	cm	0-1	%	%	#	1-5	1-5	1-5					
113	L55/CML159	105	42	30	4.31	72.6	0.3	207.6	100.2	0.52	4.9	-0.3	1.01	2.4	2.8	2.5	42.78	147.18	33.09	13.78	228.77
59	L30/CML144	105	48	25	4.28	74.3	-0.1	237.6	106.4	0.48	1.6	1.6	1.18	2.5	2.1	2.3	43.34	149.82	35.00	14.74	200.44
45	L16/CML159	104	50	32	4.27	71.3	2.0	232.0	96.6	0.44	0.0	5.8	0.96	2.7	2.4	2.2	43.02	143.09	32.40	14.52	223.30
87	L58/CML144	104	53	36	4.25	71.6	0.1	220.3	103.2	0.50	2.0	2.4	1.13	2.2	2.6	2.5	39.54	147.67	33.89	13.80	200.30
31	L2/CML159	103	47	12	4.23	72.1	1.0	224.3	93.5	0.44	0.2	10.5	0.97	2.5	2.5	2.2	44.67	147.75	34.16	14.62	226.71
54	L25/CML159	103	57	40	4.20	73.1	0.8	241.1	118.9	0.53	18.2	36.4	0.91	2.5	2.8	2.4	41.84	152.92	33.62	14.99	188.58
51	L22/CML159	103	52	36	4.19	68.8	1.6	228.6	96.6	0.46	1.8	1.8	1.04	2.4	2.6	2.3	42.32	152.32	32.64	15.00	207.27
42	L13/CML159	102	51	43	4.17	74.1	1.7	234.0	100.7	0.47	2.9	5.8	0.91	2.4	2.7	1.9	46.50	143.00	33.72	14.79	201.64
94	L36/CML159	102	58	39	4.16	71.1	-0.9	235.1	111.7	0.50	3.4	2.5	0.92	2.2	2.5	2.4	42.45	157.53	33.34	13.83	259.05
102	L44/CML159	102	52	18	4.15	73.2	1.1	227.2	93.0	0.45	1.5	6.5	1.05	2.5	2.2	2.4	41.06	155.08	35.48	14.56	185.51
28	L28/CML144	101	56	12	4.14	72.5	0.3	234.9	119.2	0.56	1.5	36.1	1.41	2.8	3.0	2.5	38.66	155.51	33.35	14.92	196.24
107	L49/CML159	101	58	35	4.14	72.4	0.1	231.2	103.5	0.49	-0.1	1.0	0.92	2.5	2.6	2.5	43.23	145.66	32.98	15.39	223.90
84	L55/CML144	101	59	30	4.13	73.0	0.2	218.4	101.3	0.50	0.2	2.3	1.12	2.2	2.5	2.5	40.36	149.48	33.25	14.23	199.45
67	L38/CML144	101	58	19	4.12	72.5	1.0	225.9	106.0	0.50	1.7	5.6	1.03	2.7	2.6	2.5	42.10	150.65	34.23	15.72	188.74
101	L43/CML159	100	59	29	4.10	73.9	1.8	225.0	98.9	0.48	0.0	5.2	0.89	2.4	2.5	2.6	42.96	158.43	35.02	14.72	204.45
83	L54/CML144	99	64	38	4.07	72.8	-0.4	224.1	106.2	0.51	1.7	5.8	1.22	2.3	2.6	2.3	39.84	151.01	33.54	13.89	194.25
37	L8/CML159	99	64	28	4.07	72.4	2.0	220.3	96.5	0.47	-0.1	2.4	1.06	2.5	2.9	2.2	43.41	141.54	31.65	13.93	225.34
56	L27/CML159	99	64	19	4.06	70.6	1.0	229.7	112.4	0.53	11.3	20.8	0.93	2.5	3.2	2.4	43.91	139.50	32.49	14.43	201.37
65	L36/CML144	99	67	47	4.06	72.3	-0.1	240.5	112.0	0.50	0.0	7.9	1.04	2.5	2.4	2.5	40.18	158.93	35.13	14.02	199.48
41	L12/CML159	99	58	41	4.06	71.5	3.1	219.1	91.4	0.43	0.1	8.4	0.95	2.5	2.8	2.1	45.24	143.80	33.40	14.93	209.49
52	L23/CML159	99	63	49	4.03	70.1	1.9	223.7	107.9	0.53	-0.1	20.7	0.95	2.4	3.1	2.3	41.43	147.57	35.40	14.00	214.92
114	L56/CML159	98	65	29	4.02	70.7	-0.4	220.8	101.9	0.49	0.1	2.3	1.01	2.1	2.8	2.5	42.31	146.19	33.42	13.91	223.72

Appendix 2. Combined data of QPM hybrids continued

Entry	Pedigree	Across			Across	Anth	ASI	Plant	Ear	Ear	Lodging		Ears/	P.sorg	Ear	Plant	ED	EL	KPR	RPE	TKW
		RelGY*	Rank		Grain Yield	Date		Height	Height	Position	Root	Stem	Plant		Aspect	Aspect					
		%	Avg*	StdDev*	t/ha	d	d	cm	cm	0-1	%	%	#	1-5	1-5	1-5					
53	L24/CML159	98	65	34	4.02	70.7	1.3	226.9	117.8	0.55	0.1	16.4	1.09	2.6	2.9	2.5	44.36	142.99	34.61	14.73	198.74
69	L40/CML144	98	65	27	4.01	72.9	-0.7	216.6	108.4	0.53	1.4	5.7	1.35	2.2	2.7	2.5	37.94	147.46	34.29	14.03	203.14
63	L34/CML144	98	65	18	4.01	74.0	0.6	227.2	111.8	0.52	0.1	2.3	1.24	2.4	2.9	2.4	40.91	156.48	34.37	13.19	188.54
55	L26/CML159	98	70	27	4.01	71.7	2.0	231.8	107.3	0.50	1.4	14.0	0.85	2.4	3.0	2.5	44.03	142.80	33.97	14.58	204.57
76	L47/CML144	98	66	55	4.00	74.3	1.6	238.1	97.9	0.44	1.8	6.8	0.97	2.3	2.0	2.5	43.04	144.81	35.86	16.47	185.73
71	L42/CML144	98	67	15	3.99	75.1	-0.8	232.5	97.2	0.45	-0.2	1.5	0.95	2.5	2.4	2.4	40.84	152.91	35.90	15.13	170.08
62	L33/CML144	97	67	34	3.97	73.1	1.4	221.5	95.8	0.47	-0.1	7.6	1.14	2.1	2.9	2.4	41.70	154.62	36.60	14.14	185.71
39	L10/CML159	97	69	37	3.95	70.1	4.3	217.0	95.9	0.47	-0.2	-0.1	1.00	2.9	2.4	2.3	45.53	140.09	31.38	14.72	234.99
19	L19/CML144	96	72	33	3.95	74.0	0.7	241.2	119.9	0.53	3.1	11.0	1.00	2.7	2.5	2.5	43.15	164.97	33.73	14.08	233.76
91	L33/CML159	96	71	19	3.93	72.3	0.8	223.1	104.8	0.49	4.7	8.7	0.97	2.1	2.8	2.5	38.90	146.98	33.68	14.60	207.55
40	L11/CML159	95	67	55	3.91	72.1	2.5	228.1	99.2	0.47	2.0	3.0	0.71	2.6	2.6	2.2	40.65	143.55	32.59	13.89	208.72
30	L1/CML159	96	70	34	3.91	71.2	2.4	236.2	99.8	0.45	-0.2	0.0	0.87	2.5	3.0	2.3	42.47	146.69	33.61	14.50	201.65
79	L50/CML144	95	72	19	3.89	73.6	-0.6	223.0	107.5	0.52	0.0	2.1	1.06	2.4	2.5	2.5	41.26	156.13	36.23	13.79	192.06
23	L23/CML144	94	79	29	3.86	72.8	0.3	232.5	121.5	0.58	0.1	42.6	1.19	2.5	3.0	2.4	39.98	152.29	35.97	14.49	178.19
21	L21/CML144	94	74	18	3.86	72.7	0.5	236.0	124.4	0.56	1.7	14.2	1.02	2.7	2.5	2.4	44.07	156.64	33.58	14.72	197.99
15	L15/CML144	94	75	24	3.84	74.6	0.6	229.4	105.5	0.49	-0.1	5.9	1.12	2.8	2.7	2.4	42.15	142.98	33.49	15.24	192.96
68	L39/CML144	94	77	23	3.83	71.6	1.2	219.5	109.9	0.54	6.9	1.2	0.97	2.5	2.8	2.2	39.66	146.65	32.32	14.39	183.40
85	L56/CML144	94	79	17	3.83	72.7	0.2	206.7	95.0	0.51	3.5	0.1	1.22	2.3	2.9	2.3	41.84	140.49	33.16	14.06	194.11
12	L12/CML144	94	76	29	3.82	71.8	1.8	207.9	96.0	0.48	-0.2	3.3	0.95	2.7	2.9	2.3	43.36	140.38	31.19	14.50	200.78
77	L48/CML144	93	78	22	3.82	73.3	1.2	232.4	103.2	0.48	0.1	12.3	1.08	2.4	2.7	2.2	41.23	147.37	35.73	14.64	178.45
74	L45/CML144	93	79	30	3.81	71.4	2.6	217.1	95.0	0.46	4.6	2.4	0.91	2.2	2.3	2.3	42.94	154.98	37.74	15.09	193.15
20	L20/CML144	93	80	13	3.81	73.4	0.7	240.8	115.6	0.51	1.6	19.8	1.07	2.6	2.8	2.5	42.82	157.13	33.64	14.26	187.56

Appendix 2. Combined data of QPM hybrids continued

Entry	Pedigree	Across			Across	Anth	ASI	Plant	Ear	Ear	Lodging		Ears/	P.sorg	Ear	Plant	ED	EL	KPR	RPE	TKW
		RelGY*	Rank		Grain Yield	Date		Height	Height	Position	Root	Stem	Plant		Aspect	Aspect					
		%	Avg*	StdDev*	t/ha	d	d	cm	cm	0-1	%	%	#	1-5	1-5	1-5					
86	L57/CML144	93	80	21	3.81	72.8	0.0	218.2	105.2	0.52	0.1	3.4	1.03	2.4	2.9	2.1	40.18	150.19	36.29	13.47	205.30
14	L14/CML144	93	80	18	3.81	73.6	1.5	230.0	111.0	0.52	0.0	12.0	1.13	2.6	2.5	2.2	40.36	146.71	34.49	14.36	173.46
36	L7/CML159	93	73	41	3.80	71.3	4.8	211.0	84.9	0.43	0.0	2.6	0.89	2.5	2.8	2.4	42.89	143.50	31.96	14.19	221.97
16	L16/CML144	93	84	19	3.79	73.5	1.4	237.5	111.2	0.50	0.2	2.6	1.08	2.5	2.7	2.1	40.35	143.60	33.12	14.41	170.87
6	L6/CML144	92	77	33	3.78	73.1	2.9	221.1	104.7	0.51	3.6	0.9	1.21	2.5	2.7	2.3	42.43	144.68	32.42	14.11	204.36
78	L49/CML144	91	73	50	3.76	73.2	-0.8	210.1	105.0	0.54	1.5	3.6	1.01	2.4	3.1	2.5	38.19	142.16	31.86	13.60	186.73
1	L1/CML144	91	84	42	3.72	73.8	0.9	230.0	103.8	0.48	3.5	3.7	1.03	2.8	2.8	2.6	43.27	146.43	32.35	15.32	182.23
7	L7/CML144	91	75	52	3.72	71.9	1.8	221.6	102.2	0.49	0.0	3.1	1.07	2.7	2.5	2.4	42.65	149.15	33.16	14.56	205.94
120	NA	91	83	30	3.70	77.7	1.3	235.8	116.1	0.52	2.6	7.5	1.38	2.4	2.9	2.5	37.81	154.09	32.90	14.31	183.21
5	L5/CML144	90	89	40	3.68	74.5	1.7	222.7	103.0	0.49	5.1	4.1	0.99	2.5	2.6	2.2	41.86	145.98	32.85	14.33	206.79
58	L29/CML159	90	84	23	3.67	71.2	0.8	224.8	116.9	0.56	3.2	30.2	0.95	2.5	3.3	2.6	42.09	144.52	33.25	14.29	195.69
18	L18/CML144	89	85	36	3.66	73.3	0.8	233.6	121.4	0.57	1.5	18.8	1.09	2.5	2.8	2.7	41.48	164.35	35.49	14.58	191.01
100	L42/CML159	90	85	34	3.66	75.0	1.2	220.7	91.4	0.45	1.8	2.2	0.94	2.5	2.5	2.5	40.28	150.53	33.45	14.19	209.07
81	L52/CML144	89	95	12	3.63	71.8	1.1	223.3	105.6	0.52	1.5	12.1	1.01	2.4	2.8	2.5	41.57	157.25	35.94	13.89	197.24
27	L27/CML144	89	92	30	3.63	72.2	1.1	223.6	117.7	0.58	7.8	37.5	1.10	2.8	3.0	2.5	35.87	149.21	33.45	14.83	172.76
25	L25/CML144	89	96	7	3.62	74.3	0.9	232.7	117.9	0.55	-0.1	26.8	1.39	2.5	2.9	2.5	40.29	151.63	34.91	15.44	174.14
46	L17/CML159	88	94	16	3.60	71.4	2.0	223.5	95.6	0.45	4.2	3.5	0.82	2.2	2.9	2.0	43.32	152.85	35.17	13.81	231.43
70	L41/CML144	88	93	13	3.60	74.1	0.9	224.6	109.3	0.52	3.3	22.8	1.07	2.5	2.7	2.1	42.36	141.37	31.50	14.53	180.76
35	L6/CML159	88	87	27	3.59	72.1	3.4	221.5	91.4	0.45	0.0	-0.2	0.88	2.6	3.0	2.2	52.28	139.50	33.35	13.88	220.52
22	L22/CML144	88	77	34	3.59	72.7	1.7	217.7	107.7	0.52	2.0	11.6	1.21	2.4	2.7	2.4	44.04	152.46	36.31	14.73	196.17
38	L9/CML159	88	79	37	3.58	71.8	2.9	213.4	93.9	0.47	1.8	6.2	0.86	2.6	2.6	2.3	41.94	142.04	32.81	14.89	208.37
8	L8/CML144	87	95	14	3.57	72.9	2.0	216.8	98.2	0.49	0.1	2.6	0.97	2.5	2.8	2.4	42.18	144.77	31.78	14.10	194.75

Appendix 2. Combined data of QPM hybrids continued

Entry	Pedigree	Across			Across	Anth	ASI	Plant	Ear	Ear	Lodging		Ears/	P.sorg	Ear	Plant	ED	EL	KPR	RPE	TKW
		RelGY*	Rank		Grain Yield	Date		Height	Height	Position	Root	Stem	Plant		Aspect	Aspect					
		%	Avg*	StdDev*	t/ha	d	d	cm	cm	0-1	%	%	#	1-5	1-5	1-5					
13	L13/CML144	87	90	30	3.55	74.0	2.9	227.0	109.7	0.52	3.2	5.5	0.89	2.4	2.4	2.5	43.14	147.55	33.34	14.60	176.98
11	L11/CML144	87	96	12	3.54	72.1	2.0	221.7	102.1	0.51	-0.1	1.2	1.06	2.3	2.8	2.4	41.48	145.28	32.09	14.39	195.21
72	L43/CML144	86	92	26	3.54	73.7	1.8	220.8	95.1	0.45	3.4	6.6	1.14	2.4	2.5	2.5	38.99	146.38	35.67	14.68	157.70
80	L51/CML144	86	95	23	3.50	71.3	-0.2	222.6	106.5	0.50	0.1	2.7	1.20	2.5	2.9	2.5	39.32	146.03	32.62	13.63	190.41
73	L44/CML144	85	96	22	3.49	74.2	0.8	225.4	92.6	0.43	0.1	0.4	1.13	2.5	2.5	2.5	43.07	147.41	36.10	15.37	167.50
24	L24/CML144	84	96	25	3.43	72.0	1.3	224.6	117.7	0.58	0.0	39.1	1.17	2.5	2.8	2.5	41.84	144.46	32.97	14.54	181.41
10	L10/CML144	80	96	31	3.25	72.1	2.6	219.0	96.1	0.46	1.5	10.4	0.86	2.5	2.7	2.4	43.30	137.51	30.78	15.04	205.43
29	L29/CML144	79	106	10	3.24	74.5	1.3	223.7	116.3	0.56	1.4	33.1	0.98	2.6	3.3	2.4	38.69	149.84	34.56	14.04	164.23
26	L26/CML144	79	98	31	3.23	73.7	2.1	224.2	109.9	0.54	4.6	28.9	0.88	2.5	3.0	2.6	42.38	136.99	33.34	14.74	176.67
9	L9/CML144	79	111	5	3.22	72.7	2.1	218.3	96.6	0.47	4.7	5.1	0.96	2.6	2.8	2.5	41.70	145.96	33.37	13.97	212.34
75	L46/CML144	76	111	9	3.08	75.1	3.5	236.2	108.7	0.49	1.4	0.7	0.95	2.4	2.9	2.6	40.86	142.53	34.55	14.52	167.36
17	L17/CML144	75	105	14	3.06	71.2	2.4	209.2	98.7	0.50	3.6	11.6	1.02	2.5	2.7	2.5	42.98	147.49	32.31	14.31	204.03
Mean		**100**	**61**	**27**	**4.09**	**72.4**	**1.2**	**227.0**	**104.0**	**0.49**	**2.1**	**8.1**	**1.03**	**2.4**	**2.7**	**2.4**	**42.11**	**149.91**	**34.01**	**14.45**	**205.47**
LSD (0.05)		**11**	**25**	**13**	**0.74**	**1.7**	**1.6**	**15.6**	**12.3**	**0.05**	**5.1**	**11.1**	**0.25**	**0.3**	**0.4**	**0.3**	**3.49**	**9.97**	**2.41**	**0.91**	**24.82**
MSe					**0.42**	**1.4**	**1.3**	**123.4**	**115.7**	**0.00**	**6.7**	**93.9**	**0.02**	**0.0**	**0.1**	**0.0**	**3.09**	**75.65**	**4.42**	**0.42**	**469.02**
Min		**75**	**11**	**5**	**3.06**	**66.7**	**-1.1**	**206.7**	**80.9**	**0.39**	**-0.2**	**-0.3**	**0.71**	**1.6**	**2.0**	**1.9**	**35.87**	**136.99**	**30.71**	**13.19**	**157.70**
Max		**131**	**111**	**63**	**5.38**	**77.7**	**4.8**	**245.6**	**124.4**	**0.58**	**18.2**	**42.6**	**1.50**	**2.9**	**3.3**	**2.7**	**52.28**	**168.07**	**37.74**	**16.47**	**259.05**
NumSignificantSites		3	3	3	3	2	2	2	3	3	1	3	1	2	3	2	1	3	3	2	3

**RelGY = Relative grain yield; Avg = Average rank; StdDev = Standard deviation of the rank*

Color Legend		
Colors that have no letter in common are different by at least one LSD. LSDs were calculated from the mean square error that was pooled across sites.	A	Very Good
	AB	Good
	BC	Average
	CD	Poor
	D	Very Poor

Appendix 3. Combined data for grain yield and other related traits in quality protein maize inbred lines evaluated at Melkassa, Edo Gojola and Mieso during 2014 main season

Entry	Stock ID	Pedigree	Across RelGY	Across Rank Avg	Across Rank StdDev	Across Grain Yield	Anth Date	ASI	Plant Height	Ear Height	Ear Position	Lodging Stem	Ears/ Plant	P.sorg	Ear Aspect	Plant Aspect	ED	EL	KPR	RPE	TKW
			%	Avg	StdDev	t/ha	d	d	cm	cm	0-1	%	#	1-5	1-5	1-5					
52	M40-54	L52	210	4	5	3.15	67.9	-0.4	153.4	68.9	0.46	3.8	1.07	2.3	2.4	2.4	39.76	138.90	28.55	12.96	216.29
38	M40-39	L38	214	3	2	2.94	63.8	2.1	162.9	52.6	0.34	4.3	0.75	3.1	1.8	2.6	40.98	125.74	28.62	13.68	218.37
47	M40-49	L47	202	3	1	2.88	68.8	1.0	163.0	60.2	0.40	7.8	0.83	2.6	2.4	2.6	41.41	127.53	27.55	15.14	205.81
17	M40-18	L17	170	13	16	2.43	64.4	1.5	167.7	71.6	0.49	8.3	0.57	2.4	2.3	2.7	41.18	123.03	29.65	13.09	225.13
48	M40-50	L48	157	6	3	2.33	69.3	-0.6	142.0	58.9	0.45	0.1	0.89	2.5	2.6	2.8	38.51	116.60	26.95	13.89	185.99
40	M40-42	L40	138	10	8	2.06	70.1	-0.7	145.2	69.0	0.50	7.5	1.24	2.4	2.9	2.9	37.58	115.76	26.90	12.46	196.91
59		CML144	138	11	7	2.02	83.9	-1.0	158.9	74.5	0.50	5.7	1.36	2.1	3.2	2.7	36.98	119.53	27.39	12.73	130.32
33	M40-34	L33	128	18	14	1.78	73.4	1.7	157.2	77.5	0.52	5.3	1.00	2.2	3.1	3.0	37.13	127.62	23.50	12.83	156.66
49	M40-51	L49	113	22	21	1.76	72.6	-1.5	138.0	65.5	0.52	3.5	0.93	2.2	3.1	3.1	35.19	104.74	22.85	12.53	191.68
25	M40-26	L25	117	16	13	1.74	71.5	0.6	162.8	75.6	0.50	25.9	1.05	2.7	3.1	2.9	35.79	111.05	23.76	13.38	148.69
36	M40-37	L36	116	15	9	1.71	71.1	-1.1	166.2	60.9	0.41	1.2	0.79	2.3	3.1	2.7	33.50	123.92	23.63	11.57	197.82
13	M40-13	L13	123	21	19	1.71	73.5	2.6	150.2	59.8	0.44	3.7	0.84	2.5	2.8	2.4	40.25	115.16	21.67	12.74	184.01
9	M40-9	L09	118	15	3	1.70	71.5	2.3	126.9	49.7	0.45	1.2	0.94	3.2	3.1	2.7	38.88	101.99	19.23	13.08	184.97
35	M40-36	L35	116	16	5	1.69	72.7	-0.5	170.9	65.1	0.41	3.1	0.87	2.0	2.9	2.7	32.70	113.02	23.09	11.53	187.69
12	M40-12	L12	115	16	3	1.68	71.3	2.6	126.4	43.6	0.41	1.0	0.92	3.0	2.7	2.6	38.86	92.84	20.68	13.89	170.12
6	M40-6	L06	117	18	10	1.67	72.6	3.4	143.0	56.2	0.42	1.9	0.81	3.0	2.6	2.5	38.07	104.05	20.65	12.55	194.12
55	M40-57	L55	109	20	21	1.65	74.0	-1.8	129.6	73.4	0.58	2.4	0.95	2.5	3.2	3.1	34.36	97.36	21.07	12.28	165.63
57	M40-59	L57	112	18	4	1.61	74.7	-1.6	132.0	64.5	0.55	-0.5	0.75	2.5	2.9	2.9	34.91	106.60	21.87	12.42	185.17
22	M40-23	L22	109	21	4	1.57	71.9	-0.2	152.0	72.4	0.49	-0.6	0.76	2.2	2.2	2.7	38.49	109.18	22.98	13.75	178.13
53	M40-55	L53	101	32	29	1.51	69.0	1.3	130.2	48.6	0.44	2.8	0.87	2.8	3.2	3.6	33.34	109.90	20.92	12.53	185.96
58	M40-60	L58	103	25	5	1.50	71.3	2.1	145.5	55.3	0.38	16.0	0.87	2.3	3.2	3.0	38.41	118.57	23.07	13.01	174.72

Appendix 3. Combined data of QPM inbred lines continued

Entry	Stock ID	Pedigree	Across RelGY	Across Rank Avg	Across Rank StdDev	Across Grain Yield	Anth Date	ASI	Plant Height	Ear Height	Ear Position	Lodging Stem	Ears/ Plant	P.sorg	Ear Aspect	Plant Aspect	ED	EL	KPR	RPE	TKW
			%	Avg	StdDev	t/ha	d	d	cm	cm	0-1	%	#	1-5	1-5	1-5					
37	M40-38	L37	102	24	6	1.50	73.1	-0.1	163.3	63.2	0.43	0.9	0.88	2.3	2.9	3.0	34.56	116.52	23.05	12.03	169.57
14	M40-15	L14	103	27	19	1.48	74.1	2.4	149.2	62.3	0.46	3.4	0.95	2.7	3.3	2.4	35.35	101.33	21.89	12.59	143.15
7	M40-7	L07	97	26	3	1.43	71.8	2.4	124.0	48.2	0.44	0.6	0.80	3.0	2.7	2.6	36.60	93.96	17.80	13.20	165.16
11	M40-11	L11	96	30	3	1.41	73.0	3.7	124.6	53.7	0.47	0.8	0.74	2.8	3.1	3.0	37.78	101.05	17.71	12.51	197.43
32	M40-33	L32	100	26	17	1.40	72.4	-1.2	136.4	50.8	0.44	16.2	0.84	2.8	2.8	3.4	36.44	111.97	21.21	12.14	196.27
56	M40-58	L56	91	30	16	1.38	72.6	-1.1	128.2	58.4	0.50	7.5	0.88	2.9	3.7	3.1	33.75	99.52	20.38	12.10	195.83
18	M40-19	L18	92	32	5	1.37	69.5	1.7	145.0	59.5	0.43	23.1	0.83	2.5	2.9	3.1	38.75	112.52	22.10	13.00	166.36
5	M40-5	L05	91	31	16	1.34	76.0	2.3	135.3	51.0	0.42	2.8	0.74	3.0	3.1	2.6	40.23	95.58	18.44	12.99	188.55
50	M40-52	L50	90	35	22	1.33	74.7	-1.5	120.7	58.7	0.54	7.4	0.83	2.5	3.2	3.3	32.88	106.51	22.19	12.59	177.04
8	M40-8	L08	89	36	1	1.31	72.9	2.8	122.3	54.4	0.46	0.5	0.80	2.8	2.8	2.6	37.45	94.81	18.05	12.86	194.85
45	M40-47	L45	88	33	7	1.30	70.7	1.8	135.5	49.1	0.41	6.4	0.79	2.8	3.7	3.2	36.33	118.89	25.95	14.18	153.17
21	M40-22	L21	88	35	9	1.29	71.6	1.1	159.2	67.7	0.47	27.7	0.88	2.7	3.9	3.2	40.14	104.37	20.74	13.10	184.81
20	M40-21	L20	95	31	22	1.29	71.3	2.5	145.4	56.0	0.44	24.0	0.74	2.8	3.1	2.7	37.57	103.48	20.27	12.81	171.94
34	M40-35	L34	91	36	24	1.29	76.4	-1.1	153.8	58.3	0.40	1.6	0.83	2.5	3.1	2.9	32.82	123.64	21.19	9.81	186.33
15	M40-16	L15	89	35	20	1.28	74.4	2.1	147.0	68.5	0.49	6.7	0.90	3.0	2.9	2.7	36.50	103.61	20.39	12.31	177.77
44	M40-46	L44	95	35	23	1.28	72.0	3.0	141.1	46.8	0.38	12.9	0.70	2.7	3.7	3.0	35.14	108.53	24.98	14.45	128.72
19	M40-20	L19	88	34	20	1.24	72.3	2.3	155.2	76.6	0.49	25.5	0.83	3.0	3.2	3.2	36.68	120.33	22.02	12.66	168.26
27	M40-28	L27	85	37	5	1.23	70.3	0.9	135.3	63.3	0.51	41.5	0.85	2.8	3.6	3.0	34.27	116.68	23.23	12.64	152.59
3	M40-3	L03	84	37	9	1.22	75.0	2.8	164.6	64.4	0.42	2.6	0.83	2.7	3.5	3.2	37.48	110.23	23.00	13.22	148.33
4	M40-4	L04	82	38	7	1.20	73.2	1.3	162.4	67.2	0.42	1.7	0.69	2.7	3.8	2.8	33.87	106.25	21.72	12.43	143.59
41	M40-43	L41	76	39	23	1.20	77.2	-2.8	142.8	67.9	0.50	0.7	0.76	2.5	3.5	3.0	34.29	105.64	21.51	12.12	177.58

Appendix 3. Combined data of QPM inbred lines continued

Entry	Stock ID	Pedigree	Across RelGY	Across Rank Avg	Across Rank StdDev	Across Grain Yield	Anth Date	ASI	Plant Height	Ear Height	Ear Position	Lodging Stem	Ears/ Plant	P.sorg	Ear Aspect	Plant Aspect	ED	EL	KPR	RPE	TKW
			%	Avg	StdDev	t/ha	d	d	cm	cm	0-1	%	#	1-5	1-5	1-5					
16	M40-17	L16	80	39	14	1.19	74.1	2.9	145.8	63.3	0.46	3.8	0.89	3.2	2.8	2.6	36.27	96.34	20.45	12.63	149.04
54	M40-56	L54	79	42	8	1.18	72.9	-2.0	133.8	59.7	0.50	1.2	0.82	2.5	3.5	3.4	33.84	96.00	21.26	12.78	177.46
28	M40-29	L28	80	40	6	1.18	70.1	0.0	140.2	66.3	0.51	34.6	0.67	3.3	3.6	3.5	31.72	110.84	22.59	12.33	139.77
51	M40-53	L51	77	42	10	1.17	74.7	-1.3	127.6	64.1	0.53	4.5	0.82	2.5	3.3	3.2	33.59	100.90	20.12	12.27	172.04
29	M40-30	L29	72	43	17	1.13	70.6	0.5	138.0	71.7	0.53	36.6	0.81	2.8	3.7	3.2	34.89	110.40	23.06	12.07	158.53
23	M40-24	L23	79	39	13	1.12	71.0	1.4	139.3	66.4	0.49	27.4	0.84	2.8	3.1	3.5	32.63	104.81	23.36	13.55	131.42
60		CML159	68	44	22	1.07	81.3	1.0	138.4	53.5	0.44	2.0	0.73	2.0	4.0	3.0	36.15	125.04	21.56	13.29	183.50
39	M40-41	L39	71	45	10	1.05	70.2	-1.4	134.4	42.8	0.36	5.5	0.71	3.0	2.9	3.4	35.32	80.66	20.19	13.07	160.76
42	M40-44	L42	69	45	15	1.02	71.2	1.9	135.7	52.1	0.42	13.3	0.76	2.8	3.2	3.1	47.38	82.89	14.57	13.82	200.46
31	M40-32	L31	73	43	13	1.02	71.6	1.6	134.8	63.9	0.53	20.4	1.00	2.6	3.5	3.8	34.00	111.99	22.07	12.29	159.56
2	M40-2	L02	70	45	11	0.98	73.8	0.9	157.7	62.0	0.41	3.2	0.68	3.2	4.0	3.3	34.37	100.30	21.01	13.38	146.32
43	M40-45	L43	65	48	8	0.94	74.7	3.3	143.5	54.2	0.39	9.2	0.73	2.7	3.5	3.1	35.30	118.10	24.49	13.24	154.41
10	M40-10	L10	62	50	8	0.93	72.2	3.8	112.0	44.9	0.45	0.8	0.80	3.0	2.9	3.4	36.22	79.68	15.27	13.12	177.36
1	M40-1	L01	64	50	5	0.92	70.8	2.4	147.8	50.4	0.39	-0.5	0.79	2.8	3.7	3.0	34.84	95.58	18.19	13.24	169.52
26	M40-27	L26	62	50	9	0.91	71.4	2.7	136.0	70.2	0.51	35.9	0.62	3.0	3.6	3.6	37.94	93.43	21.51	14.12	128.23
30	M40-31	L30	61	50	6	0.88	76.7	0.5	121.8	60.9	0.50	5.9	0.72	2.7	4.1	3.4	35.57	97.96	19.86	13.11	157.66
24	M40-25	L24	50	48	21	0.75	70.7	2.0	138.8	64.3	0.49	40.7	0.74	3.0	3.5	3.6	33.08	97.19	19.95	12.88	139.28
46	M40-48	L46	48	54	.	0.54	75.1	2.0	141.3	45.6	0.36	-0.5	.	2.2	3.2	2.9	37.67	92.16	19.55	14.40	167.42
Mean			**99**	**30**	**11**	**1.45**	**72.4**	**1.0**	**143.0**	**60.4**	**0.46**	**9.4**	**0.83**	**2.7**	**3.2**	**3.0**	**36.43**	**107.55**	**22.02**	**12.89**	**172.40**
LSD (0.05)			**35**	**13**	**7**	**0.45**	**1.4**	**1.6**	**11.5**	**9.3**	**0.06**	**8.8**	**0.22**	**0.6**	**0.5**	**0.4**	**4.05**	**9.33**	**2.13**	**0.84**	**21.89**
MSe						**0.15**	**0.9**	**1.3**	**97.5**	**42.3**	**0.00**	**56.1**	**0.02**	**0.1**	**0.1**	**0.1**	**7.99**	**63.59**	**3.32**	**0.51**	**350.29**
Min			**48**	**3**	**1**	**0.54**	**63.8**	**-2.8**	**112.0**	**42.8**	**0.34**	**-0.6**	**0.57**	**2.0**	**1.8**	**2.4**	**31.72**	**79.68**	**14.57**	**9.81**	**128.23**
Max			**214**	**54**	**29**	**3.15**	**83.9**	**3.8**	**170.9**	**77.5**	**0.58**	**41.5**	**1.36**	**3.3**	**4.1**	**3.8**	**47.38**	**138.90**	**29.65**	**15.14**	**225.13**
NumSignificantSites			3	3	3	3	2	2	3	2	3	3	2	1	2	2	2	3	3	3	3

Appendix 4. Percentage mid-parent heterosis of QPM hybrids evaluated at Melkassa, Edo Gojola and Mieso during 2014 main season

Hybrids	GY		DA		ASI		PH		EH		EPO		SL		CLR	EA		PA		EL		KPR		RPE		TKW	
L1/CML144	153.2	**	-4.6	**	39.3		50.0	**	66.3	**	8.1		44.0		14.2	-18.5	*	-9.2		36.1	**	42.0	**	18.0	**	21.5	
L2/CML144	203.0	**	-7.4	**	-467.1		54.2	**	53.9	**	2.5		1.5		-11.1	-29.9	**	-24.8	**	37.4	**	45.0	**	16.6	**	37.9	**
L3/CML144	177.3	**	-8.1	**	-33.1		49.2	**	42.2	**	-4.4		2.9		-0.8	-25.9	**	-28.6	**	31.2	**	35.4	**	16.1	**	48.0	**
L4/CML144	190.6	**	-5.4	**	-316.1		45.8	**	55.9	**	20.1	*	63.1		-2.0	-26.7	**	-19.1	**	37.7	**	44.0	**	13.9	**	49.5	**
L5/CML144	118.9	**	-6.8	**	178.9		51.4	**	64.1	**	7.6		-4.5		-2.2	-18.0		-15.8	*	35.7	**	43.4	**	11.5	**	29.7	*
L6/CML144	104.5	**	-6.6	**	144.3		46.5	**	60.2	**	9.7		-75.0		-2.3	-6.8		-13.1		29.4	**	35.0	**	11.6	**	26.0	*
L7/CML144	116.0	**	-7.6	**	160.3		56.6	**	66.6	**	5.1		-2.2		8.7	-14.9		-12.2		39.7	**	46.8	**	12.3	**	39.4	**
L8/CML144	114.7	**	-7.0	**	133.4		54.2	**	52.4	**	2.5		-15.2		3.8	-7.5		-10.7		35.1	**	39.9	**	10.2	*	19.8	
L9/CML144	73.1	*	-6.4	**	226.8		52.7	**	55.6	**	-1.4		47.6		-2.1	-9.4		-9.8		31.8	**	43.1	**	8.2		34.7	**
L10/CML144	120.4	**	-7.5	**	88.2		61.7	**	60.9	**	-2.8		216.4		-2.3	-11.9		-22.7	**	38.1	**	44.3	**	16.4	**	33.5	**
L11/CML144	106.7	**	-8.0	**	47.4		56.4	**	59.4	**	4.9		-63.7		-7.0	-11.6		-17.7	**	31.7	**	42.3	**	14.0	**	19.1	
L12/CML144	107.0	**	-7.5	**	132.2		45.7	**	62.5	**	5.6		-0.2		8.2	0.6		-12.9		32.2	**	29.8	**	9.0	*	33.7	**
L13/CML144	90.2	**	-6.0	**	278.4	*	46.9	**	63.4	**	10.4		16.6		2.7	-19.5		-3.1		25.7	**	35.9	**	14.6	**	12.6	
L14/CML144	117.8	**	-6.8	**	115.5		49.3	**	62.2	**	7.7		164.4		9.1	-21.8	*	-15.9	*	32.9	**	40.0	**	13.4	**	26.9	
L15/CML144	132.7	**	-5.8	**			50.0	**	47.5	**	-1.1		-5.2		9.3	-12.6		-11.9		28.2	**	40.2	**	21.7	**	25.3	*
L16/CML144	136.2	**	-6.9	**			55.9	**	61.4	**	5.0		-44.6		-4.8	-11.1		-21.0	**	33.0	**	38.5	**	13.6	**	22.3	
L17/CML144	37.5		-3.9	**	1.6		28.1	**	35.2	**	2.5		66.4		10.5	-3.7		-7.8		21.6	**	13.3	*	10.8	*	14.8	
L18/CML144	116.5	**	-4.4	**	54.5		53.7	**	81.2	**	22.0	**	30.6		8.9	-8.9		-7.5		41.6	**	43.4	**	13.3	**	28.8	*
L19/CML144	141.8	**	-5.3	**	17.2		53.6	**	58.7	**	8.5		-29.6		7.0	-20.9	*	-15.6	*	37.6	**	36.6	**	10.9	*	56.6	**
L20/CML144	130.3	**	-5.5	**	-4.6		58.3	**	77.1	**	8.7		33.7		7.8	-9.3		-7.8		40.9	**	41.2	**	11.6	**	24.1	
L21/CML144	132.9	**	-6.4	**	1243.1		48.3	**	75.1	**	15.7		-15.3		13.6	-31.0	**	-19.9	**	39.9	**	39.5	**	14.0	**	25.7	*
L22/CML144	100.1	**	-6.7	**	-369.2	*	40.1	**	46.6	**	5.8		350.8		9.6	-2.0		-13.0		33.3	**	44.2	**	11.3	**	27.2	*
L23/CML144	146.0	**	-6.0	**	67.7		56.0	**	72.5	**	17.8	*	157.5	**	3.2	-4.6		-22.1	**	35.8	**	41.8	**	10.3	*	36.2	*
L24/CML144	147.7	**	-6.9	**	166.7		50.9	**	69.6	**	17.4	*	68.6		-2.3	-15.6		-20.7	**	33.3	**	39.3	**	13.6	**	34.6	*
L25/CML144	92.6	**	-4.4	**	-523.7		44.7	**	57.0	**	10.3		69.6		4.5	-6.4		-11.7		31.5	**	36.5	**	18.3	**	24.8	
L26/CML144	120.8	**	-5.1	**	150.9		52.0	**	51.8	**	6.7		38.9		-1.6	-13.0		-17.8	**	28.7	**	36.4	**	9.8	*	36.7	*
L27/CML144	123.5	**	-6.4	**	-1238.5		52.0	**	70.7	**	13.8		59.1		12.8	-11.0		-13.8	*	26.3	**	32.2	**	16.9	**	22.1	
L28/CML144	159.5	**	-5.8	**	-167.6		57.1	**	69.3	**	10.6		79.5		2.8	-11.2		-21.0	**	35.0	**	33.4	**	19.1	**	45.3	**
L29/CML144	105.7	**	-3.5	**	-623.8		50.7	**	59.1	**	9.1		56.5		7.8	-3.5		-20.3	**	30.3	**	37.0	**	13.2	**	13.7	
L30/CML144	195.5	**	-7.4	**	-55.8		69.3	**	57.1	**	-3.2		-72.2		4.3	-42.1	**	-26.9	**	37.8	**	48.1	**	14.1	**	39.2	**

Appendix 4. Percentage mid-parent heterosis of QPM hybrids continued

Hybrids	GY		DA		ASI		PH		EH		EPO		SL		CLR		EA		PA		EL		KPR		RPE		TKW	
L31/CML144	184.6	**	-7.7	**	64.7		53.4	**	47.0	**	-5.7		-82.1		2.7		-33.0	**	-21.0	**	32.3	**	50.0	**	31.1	**	46.6	**
L32/CML144	173.1	**	-6.1	**	2.6		60.0	**	77.1	**	7.6		-65.4		-1.9		-29.8	**	-14.4	*	34.3	**	45.4	**	16.7	**	26.7	*
L33/CML144	109.3	**	-7.1	**	332.6		40.1	**	26.1	*	-7.7		36.9		-0.3		-8.3		-17.3	**	25.1	**	43.8	**	10.6	*	29.4	*
L34/CML144	143.0	**	-7.6	**	-158.8		45.3	**	68.5	**	15.5		-38.1		4.7		-8.8		-15.1	*	28.7	**	41.5	**	17.0	**	19.1	
L35/CML144	149.2	**	-6.2	**	30.6		46.1	**	66.2	**	13.5		12.4		23.3	*	-22.8	*	-21.6	**	40.7	**	42.8	**	19.2	**	20.6	
L36/CML144	117.8	**	-6.6	**	-92.7		48.0	**	65.4	**	11.9		128.5		15.2		-23.7	*	-9.1		30.6	**	37.7	**	15.4	**	21.6	
L37/CML144	149.7	**	-6.2	**	-171.4		50.5	**	59.6	**	5.3		117.1		9.2		-22.7	*	-20.0	**	30.3	**	39.0	**	14.3	**	32.4	*
L38/CML144	66.3	**	-1.7		81.9		40.4	**	66.8	**	20.5	*	11.1		2.7		4.6		-6.9		22.8	**	22.2	**	19.0	**	8.3	
L39/CML144	150.3	**	-7.1	**	-196.8	*	49.7	**	87.4	**	25.0	**	-78.5		-1.6		-7.4		-26.7	**	46.5	**	35.9	**	11.5	**	26.0	*
L40/CML144	96.8	**	-5.3	**	-13.2		42.5	**	51.1	**	6.6		-13.6		-0.8		-11.2		-12.4		25.3	**	26.3	**	11.4	*	24.2	*
L41/CML144	124.1	**	-8.0	**	-145.0	**	48.9	**	53.5	**	3.7		607.5	*	8.5		-20.3	*	-24.9	**	25.6	**	28.8	**	16.9	**	17.4	
L42/CML144	162.3	**	-3.1	*	-277.3		57.9	**	53.6	**	-2.1		-84.3		3.0		-25.1	*	-18.6	**	51.1	**	71.1	**	14.0	**	2.8	
L43/CML144	139.1	**	-7.0	**	60.3		46.0	**	47.8	**	3.0		-10.9		-1.6		-23.9	*	-14.6	*	23.2	**	37.5	**	13.1	**	10.8	
L44/CML144	111.2	**	-4.8	**	-19.1		50.3	**	52.6	**	-1.2		-95.6		3.3		-27.2	**	-13.0		29.3	**	37.9	**	13.1	**	29.3	*
L45/CML144	129.6	**	-7.6	**	577.0	*	47.5	**	53.7	**	1.2		-59.6		-7.7		-32.6	**	-20.4	**	30.0	**	41.5	**	12.1	**	36.3	**
L46/CML144	141.3	**	-5.5	**	593.2	**	57.4	**	81.1	**	14.3		-72.5		9.7		-8.9		-6.2		34.7	**	47.2	**	7.0		12.4	
L47/CML144	63.2	**	-2.7	*	122221.1		47.9	**	45.4	**	-1.6		1.3		-2.6		-27.7	*	-6.6		17.2	**	30.5	**	18.2	**	10.5	
L48/CML144	75.6	**	-4.3	**	-241.3	*	54.5	**	54.7	**	1.5		325.1		4.0		-7.1		-18.1	**	24.8	**	31.5	**	10.0	*	12.8	
L49/CML144	98.8	**	-6.4	**	-39.9		41.6	**	49.9	**	5.7		-22.0		10.4		-3.6		-14.5	*	26.8	**	26.8	**	7.7		16.0	
L50/CML144	132.5	**	-7.1	**	-56.7		59.5	**	61.5	**	0.7		-68.4		4.0		-22.4	*	-17.5	**	38.1	**	46.1	**	8.9	*	25.0	*
L51/CML144	120.0	**	-10.0	**	-85.0		55.4	**	53.6	**	-1.9		-47.2		8.2		-10.2		-16.5	*	32.5	**	37.3	**	9.1	*	26.0	*
L52/CML144	40.5		-5.4	**	-259.5		43.0	**	47.3	**	8.2		155.1		9.1		0.9		-2.9		21.7	**	28.5	**	8.2		13.8	
L53/CML144	158.8	**	-6.2	**	494.9		56.0	**	91.8	**	18.6	*	84.5		1.1		-15.4		-24.7	**	40.3	**	47.9	**	14.6	**	26.5	*
L54/CML144	154.1	**	-7.1	**	-76.5		53.1	**	58.3	**	2.2		68.0		-1.6		-22.1	*	-26.7	**	40.1	**	37.9	**	8.9	*	26.2	*
L55/CML144	125.4	**	-7.5	**	-111.1		51.4	**	37.1	**	-6.8		-43.2		-2.2		-21.2	*	-14.4	*	37.8	**	37.2	**	13.8	**	34.8	**
L56/CML144	125.7	**	-7.0	**	-121.1		44.0	**	43.1	**	1.5		-97.9		-9.8		-16.9		-22.8	**	28.3	**	38.8	**	13.3	**	19.0	
L57/CML144	109.8	**	-8.2	**	-102.9		50.0	**	51.4	**	-0.5		30.7		3.7		-7.4		-24.0	**	32.8	**	47.3	**	7.1		30.1	*
L58/CML144	141.5	**	-7.7	**	-74.9		44.8	**	59.1	**	15.1		-77.5		1.8		-19.3		-12.9		24.0	**	34.3	**	7.2		31.3	*
L1/CML159	292.3	**	-6.3	**	42.9		65.0	**	92.2	**	8.8		-104.5		5.2		-20.3	*	-22.2	**	33.0	**	69.1	**	9.3	*	14.2	
L2/CML159	312.7	**	-7.0	**	-2.1		51.5	**	61.8	**	3.4		307.2		-4.4		-37.5	**	-31.2	**	31.1	**	60.5	**	9.6	*	37.5	**

Appendix 4. Percentage mid-parent heterosis of QPM hybrids continued

Hybrids	GY		DA		ASI		PH		EH		EPO		SL		CLR	EA		PA		EL		KPR		RPE		TKW	
L3/CML159	285.3	**	-8.6	**	2.2		60.2	**	56.8	**	-1.4		-79.5		12.7	-22.3	**	-31.6	**	28.7	**	51.8	**	8.0		29.9	**
L4/CML159	289.3	**	-7.0	**	38.8		51.7	**	61.2	**	6.0		130.3		5.6	-26.2	**	-30.7	**	27.1	**	68.6	**	14.1	**	24.7	*
L5/CML159	283.1	**	-8.0	**	30.5		65.9	**	89.9	**	8.7		36.6		0.2	-34.1	**	-15.6	*	34.1	**	63.3	**	8.0		29.1	**
L6/CML159	161.3	**	-6.3	**	51.5		57.4	**	66.6	**	3.5		-110.1		4.7	-8.3		-17.9	*	21.8	**	58.1	**	7.4		16.8	
L7/CML159	204.4	**	-6.8	**	174.7	**	60.8	**	66.8	**	-2.1		105.9		1.2	-15.3		-15.7	*	31.1	**	62.4	**	7.1		27.3	*
L8/CML159	241.7	**	-6.1	**	6.2		69.0	**	79.0	**	4.2		94.2		4.9	-15.3		-20.0	**	28.8	**	59.8	**	6.5		19.1	
L9/CML159	158.0	**	-6.0	**	75.7		60.9	**	81.9	**	5.3		294.3		0.4	-26.4	**	-17.9	**	25.1	**	60.9	**	12.9	**	13.1	
L10/CML159	294.2	**	-8.6	**	77.4		73.4	**	94.9	**	6.4		-105.8		14.4	-31.0	**	-27.7	**	36.9	**	70.4	**	11.5	**	30.2	**
L11/CML159	215.0	**	-6.5	**	5.3		73.5	**	85.1	**	4.0		121.4		9.5	-27.7	**	-24.9	**	27.0	**	66.0	**	7.6		9.6	
L12/CML159	195.0	**	-6.3	**	69.0		65.5	**	88.2	**	3.0		473.1		0.2	-15.3		-24.5	**	32.0	**	58.1	**	9.9	*	18.5	
L13/CML159	199.2	**	-4.3	**	-7.2		62.2	**	77.8	**	6.0		105.5		3.7	-21.3	*	-30.8	**	19.1	**	56.0	**	13.6	**	9.7	
L14/CML159	240.5	**	-5.4	**	18.0		58.5	**	77.5	**	3.7		-18.3		10.5	-34.8	**	-21.5	**	28.9	**	53.1	**	6.8		36.3	**
L15/CML159	266.5	**	-5.1	**	126.2	*	61.4	**	66.6	**	3.4		-92.6		-4.2	-23.6	*	-24.6	**	24.0	**	58.8	**	16.7	**	11.3	
L16/CML159	277.4	**	-8.2	**	0.2		63.3	**	65.4	**	-1.0		99.0		5.3	-29.2	**	-19.5	**	29.3	**	54.3	**	12.0	**	34.3	**
L17/CML159	106.0	**	-1.9		58.1		46.0	**	52.9	**	-1.9		-32.3		1.3	-8.1		-28.6	**	23.2	**	37.4	**	4.7		13.3	
L18/CML159	315.5	**	-4.8	**	-5.4		69.2	**	92.7	**	13.9		-65.9		12.1	-40.4	**	-26.7	**	30.2	**	56.7	**	9.3	*	17.9	
L19/CML159	283.4	**	-4.5	**	22.0		65.5	**	64.2	**	2.0		-37.9		-0.5	-23.0	*	-17.4	**	37.0	**	57.0	**	14.8	**	24.8	*
L20/CML159	280.7	**	-6.6	**	16.4		63.0	**	91.7	**	9.5		17.6		3.8	-37.9	**	-16.9	*	36.0	**	63.5	**	8.0		27.3	**
L21/CML159	288.3	**	-6.3	**	-48.8		63.7	**	77.5	**	4.6		-14.1		5.9	-31.9	**	-24.7	**	43.7	**	63.5	**	12.9	**	15.0	
L22/CML159	217.7	**	-10.2	**	269.6		57.5	**	53.5	**	-1.5		159.1		12.9	-15.1		-19.3	**	30.1	**	46.5	**	10.9	**	14.6	
L23/CML159	267.0	**	-7.9	**	60.0		61.2	**	80.0	**	15.1		41.3		-0.5	-10.6		-29.4	**	28.4	**	57.7	**	4.3		36.5	**
L24/CML159	341.2	**	-6.9	**	-13.9		63.7	**	100.0	**	18.9	*	-23.3		5.1	-23.8	**	-25.1	**	28.7	**	66.8	**	12.6	**	23.1	*
L25/CML159	198.3	**	-4.3	**	-9.4		60.1	**	84.2	**	14.2		161.3	**	5.0	-21.5	*	-20.0	**	29.5	**	48.4	**	12.4	**	13.5	
L26/CML159	304.7	**	-6.1	**	4.2		69.0	**	73.4	**	4.5		-25.9		-4.8	-20.2	*	-23.3	**	30.7	**	57.7	**	6.4		31.2	**
L27/CML159	252.9	**	-6.9	**	2.0		67.9	**	92.4	**	11.1		-4.1		4.4	-16.9	*	-20.7	**	15.4	*	45.1	**	11.3	**	19.8	
L28/CML159	286.3	**	-6.0	**	-112.5		67.9	**	90.5	**	10.7		-10.7		-5.3	-20.1	*	-23.5	**	33.0	**	57.4	**	9.0	*	33.0	**
L29/CML159	233.6	**	-6.2	**	-4.1		62.7	**	86.8	**	15.6		56.7		4.5	-12.4		-15.2	*	22.8	**	49.1	**	12.7	**	14.4	
L30/CML159	352.7	**	-9.9	**	-4.7		72.0	**	74.1	**	2.0		-55.4		5.3	-41.8	**	-29.0	**	30.8	**	58.4	**	8.7	*	27.2	*
L31/CML159	337.8	**	-7.2	**	-21.2		68.2	**	67.4	**	-3.9		-56.7		-7.3	-29.1	**	-29.5	**	18.8	**	59.7	**	15.2	**	28.0	*
L32/CML159	259.0	**	-7.7	**	-510.1		65.6	**	102.3	**	12.5		-27.7		-5.6	-26.9	**	-21.7	**	27.1	**	60.3	**	9.4	*	18.7	

Appendix 4. Percentage mid-parent heterosis of QPM hybrids continued

Hybrids	GY		DA		ASI		PH		EH		EPO		SL	CLR	EA		PA		EL		KPR		RPE		TKW	
L33/CML159	175.6	**	-6.5	**	-38.6		50.9	**	60.0	**	3.3		139.2	0.1	-19.7	*	-15.5	*	16.3	**	49.5	**	11.8	**	22.0	*
L34/CML159	309.5	**	-8.7	**	1464.3		61.5	**	93.2	**	17.8		-60.5	12.5	-22.3	*	-14.4	*	33.5	**	65.8	**	14.3	**	30.4	**
L35/CML159	263.2	**	-6.0	**	5.6		51.1	**	87.0	**	17.6		64.2	0.3	-18.4	*	-27.2	**	38.0	**	54.5	**	15.0	**	29.5	**
L36/CML159	198.6	**	-6.6	**	6842.7		54.4	**	95.4	**	19.0	*	58.8	5.5	-28.5	**	-15.3	*	26.6	**	47.6	**	11.2	*	35.9	**
L37/CML159	280.4	**	-5.5	**	-76.6		62.8	**	94.4	**	16.0		45.4	11.8	-33.4	**	-24.5	**	30.7	**	50.4	**	9.7	*	28.2	**
L38/CML159	126.8	**	-5.4	**	-2.3		44.5	**	52.5	**	0.6		229.8	-1.2	2.7		-10.8		21.4	**	44.2	**	10.5	*	4.8	
L39/CML159	333.1	**	-7.8	**	-347.8		68.3	**	112.3	**	20.8	*	13.9	-4.3	-25.2	**	-16.8	**	48.2	**	71.1	**	5.7		39.1	**
L40/CML159	188.9	**	-5.8	**	-131.6		60.2	**	85.1	**	16.0		-69.7	1.9	-22.7	*	-13.4	*	26.0	**	46.4	**	3.3		20.4	*
L41/CML159	301.1	**	-9.2	**	-434.4	**	66.0	**	87.9	**	11.7		449.0	4.7	-40.3	**	-28.8	**	28.1	**	53.1	**	20.1	**	23.5	*
L42/CML159	249.1	**	-1.6		-17.2		61.1	**	73.3	**	4.4		-71.2	4.2	-30.4	**	-18.1	**	44.8	**	85.2	**	4.7		8.9	
L43/CML159	307.0	**	-5.3	**	-18.1		59.7	**	83.7	**	15.6		-6.8	0.0	-33.0	**	-13.7	*	30.3	**	52.1	**	11.0	*	21.0	
L44/CML159	252.7	**	-4.5	**	-47.0		62.6	**	85.4	**	9.7		-12.2	5.5	-41.8	**	-20.2	**	32.8	**	52.5	**	4.9		18.8	
L45/CML159	310.8	**	-6.2	**	18.3		66.6	**	62.4	**	-6.7		10.1	-1.4	-43.3	**	-22.7	**	23.8	**	51.3	**	11.1	**	23.2	*
L46/CML159	437.5	**	-4.6	**	79.7		67.9	**	83.5	**	1.5		710.7	11.2	-30.0	**	-16.2	*	36.6	**	73.6	**	7.2		19.0	
L47/CML159	119.3	**	-3.2	*	149.3		48.8	**	70.4	**	11.4		117.5	9.5	-22.4	*	-5.1		15.5	**	40.0	**	1.3		12.9	
L48/CML159	160.5	**	-2.5		660.6		48.6	**	51.9	**	-4.2		1394.4	10.7	-16.9		-13.0		23.4	**	38.1	**	12.5	**	5.1	
L49/CML159	192.4	**	-5.9	**	-161.0		67.4	**	73.8	**	1.3		-62.0	18.1	* -26.2	**	-18.3	**	26.8	**	48.6	**	19.2	**	19.4	
L50/CML159	274.9	**	-8.7	**	-9.3		67.4	**	73.0	**	-1.9		1.3	11.8	-23.8	**	-24.8	**	36.2	**	58.1	**	8.2		27.7	**
L51/CML159	295.2	**	-8.8	**	-558.0		68.3	**	72.2	**	1.0		-34.4	-5.8	-34.8	**	-18.5	**	34.4	**	67.1	**	9.4	*	28.7	**
L52/CML159	154.7	**	-5.2	**	-223.0		57.3	**	54.6	**	-3.0		-69.9	9.8	-24.9	*	-6.3		23.0	**	42.0	**	12.5	**	17.3	
L53/CML159	275.2	**	-5.9	**	77.8		73.9	**	104.5	**	12.2		308.3	3.1	-28.7	**	-24.2	**	30.1	**	44.6	**	9.4	*	27.8	**
L54/CML159	303.2	**	-8.6	**	-49.5		63.8	**	78.3	**	3.7		237.7	17.0	* -34.0	**	-21.9	**	36.7	**	53.0	**	8.4		33.1	**
L55/CML159	216.3	**	-6.5	**	-170.2		55.0	**	57.9	**	2.2		-112.5	6.1	-22.9	*	-18.0	**	32.4	**	55.3	**	7.7		31.1	**
L56/CML159	228.5	**	-8.1	**	750.7		65.7	**	82.1	**	4.4		-51.0	-14.5	-25.7	**	-18.1	**	30.2	**	59.4	**	9.6	*	18.0	
L57/CML159	262.1	**	-8.5	**	-193.3		61.1	**	74.9	**	3.6		211.3	5.4	-34.4	**	-19.6	**	32.9	**	57.1	**	7.4		33.5	**
L58/CML159	262.8	**	-6.7	**	-111.3		55.2	**	83.3	**	18.2		-91.1	15.8	-39.5	**	-15.9	*	20.3	**	48.8	**	1.8		33.1	**

Appendix 5. Percentage better parent heterosis of QPM hybrids evaluated at Melkassa, Edo Gojola and Mieso during 2014 main season

Hybrids	GY		DA		ASI		PH		EH		EPO	SL	CLR		EA		PA		EL		KPR		RPE		TKW	
L1/CML144	84.4	**	-12.1	**	-60.8		44.8	**	39.4	**	-3.3	-34.4	0.3		-23.5	*	-13.7		22.5	**	18.1	*	15.8	**	7.5	
L2/CML144	124.8	**	-13.0	**	-74.2		53.6	**	41.0	**	-6.5	-20.9	-26.8	**	-36.7	**	-31.5	**	26.4	**	28.1	**	13.8	**	30.4	*
L3/CML144	122.3	**	-13.0	**	-78.9	*	46.6	**	32.6	*	-11.7	-25.4	-11.9		-28.7	**	-34.2	**	26.1	**	24.5	**	14.0	**	39.0	**
L4/CML144	131.5	**	-11.4	**	-123.8		44.2	**	48.2	**	10.7	5.9	-13.2		-32.3	**	-20.4	**	30.0	**	29.1	**	12.5	*	42.6	**
L5/CML144	82.1	*	-11.2	**	-24.2		40.2	**	38.3	**	-0.4	-28.5	-17.2	*	-19.7		-18.1	*	22.1	**	19.9	**	10.4	*	9.7	
L6/CML144	87.1	**	-12.8	**	-14.8		39.2	**	40.6	**	1.8	-83.4	-17.1	*	-15.6		-16.4	*	21.0	**	18.3	*	10.8	*	5.3	
L7/CML144	84.3	**	-14.3	**	-25.6		39.5	**	37.3	*	-0.8	-46.3	-7.1		-21.1		-13.9		24.8	**	21.1	**	10.2	*	24.7	
L8/CML144	76.9	*	-13.1	**	-27.4		36.5	**	31.8	*	-0.9	-53.6	-8.7		-13.8		-13.0		21.1	**	16.0	*	9.6		-0.1	
L9/CML144	59.5		-13.3	**	-10.1		37.4	**	29.7	*	-5.6	-10.9	-19.4	**	-11.4		-9.8		22.1	**	21.8	**	6.8		14.8	
L10/CML144	61.0		-14.0	**	-31.5		37.9	**	29.0	*	-7.0	81.6	-17.5	*	-15.6		-30.2	**	15.0	*	12.4		14.7	**	15.8	
L11/CML144	75.6	*	-14.0	**	-46.9		39.5	**	37.1	*	2.0	-79.4	-18.6	*	-13.0		-21.6	**	21.5	**	17.2	*	13.0	*	-1.1	
L12/CML144	89.5	**	-14.4	**	-30.0		30.9	**	28.9	*	-3.6	-41.6	-7.9		-8.2		-14.5		17.4	*	13.9		4.4		18.0	
L13/CML144	75.8	*	-11.8	**	13.1		42.9	**	47.3	**	4.5	-4.3	-6.3		-24.5	*	-8.2		23.4	**	21.7	**	14.6	**	-3.8	
L14/CML144	88.5	**	-12.2	**	-38.9		44.8	**	48.9	**	4.1	110.7	-3.5		-22.4	*	-20.8	*	22.7	**	25.9	**	12.8	*	21.2	
L15/CML144	90.4	**	-11.1	**	-73.9		44.4	**	41.6	**	-1.4	-12.1	-6.7		-16.4		-13.2		19.6	**	22.3	**	19.7	**	8.5	
L16/CML144	87.7	**	-12.3	**	-50.5		49.5	**	49.3	**	1.1	-53.6	-21.4	**	-17.0		-22.6	**	20.1	**	20.9	**	13.2	*	14.6	
L17/CML144	26.0		-15.1	**	59.0		24.7	**	32.5	*	1.8	40.5	2.8		-17.0		-8.9		19.9	**	9.0		9.3		-9.4	
L18/CML144	81.5	*	-12.6	**	-53.9		47.0	**	63.0	**	14.4	-18.6	0.0		-12.7		-12.6		37.5	**	29.6	**	12.2	*	14.8	
L19/CML144	95.5	**	-11.8	**	-68.1		51.8	**	56.5	**	7.7	-56.9	-9.7		-21.0		-21.5	**	37.1	**	23.2	**	10.6	*	38.9	**
L20/CML144	88.8	**	-12.5	**	-72.0		51.6	**	55.2	**	2.2	-17.2	-5.6		-11.4		-7.8		31.5	**	22.8	**	11.3	*	9.1	
L21/CML144	91.1	**	-13.3	**	-51.5		48.2	**	67.1	**	12.9	-48.9	0.5		-37.2	**	-25.3	**	31.0	**	22.6	**	12.3	*	7.1	
L22/CML144	77.7	*	-13.4	**	-930.7		37.1	**	44.6	**	5.0	103.3	6.7		-16.8		-13.2		27.5	**	32.6	**	7.1		10.1	
L23/CML144	91.4	**	-13.2	**	-79.3		46.4	**	63.2	**	16.6	55.5	-9.3		-6.8		-30.6	**	27.4	**	31.3	**	7.0		35.6	*
L24/CML144	69.8	*	-14.2	**	-36.4		41.4	**	58.1	**	16.7	-3.9	-17.2	*	-19.4		-30.7	**	20.9	**	20.4	**	12.9	*	30.2	
L25/CML144	79.5	*	-11.4	**	36.6		43.0	**	55.9	**	10.0	3.5	-7.7		-8.7		-14.8	*	26.9	**	27.4	**	15.4	**	17.1	
L26/CML144	60.0		-12.1	**	-22.3		41.1	**	47.5	**	4.8	-19.5	-16.4	*	-17.4		-28.1	**	14.6	*	21.7	**	4.4		35.6	*
L27/CML144	79.9	*	-13.9	**	25.5		40.7	**	57.9	**	11.7	-9.5	-1.2		-16.3		-18.2	*	24.8	**	22.1	**	16.5	**	13.2	
L28/CML144	105.3	**	-13.6	**	4036.5		47.9	**	60.0	**	8.8	4.5	-15.6	*	-16.5		-30.0	**	30.1	**	21.8	**	17.2	**	40.4	**
L29/CML144	60.3		-11.2	**	136.6		40.8	**	56.1	**	6.0	-9.6	-5.2		-9.6		-26.3	**	25.4	**	26.2	**	10.2	*	3.6	
L30/CML144	112.2	**	-11.4	**	-123.0		49.6	**	42.8	**	-3.6	-72.7	-8.0		-48.4	**	-34.5	**	25.3	**	27.8	**	12.5	*	27.1	*

Appendix 5. Percentage better parent heterosis of QPM hybrids continued

Hybrids	GY		DA		ASI		PH		EH		EPO	SL	CLR		EA		PA		EL		KPR		RPE		TKW	
L31/CML144	113.9	**	-14.5	**	-71.3		41.8	**	36.6	*	-8.4	-88.6	-7.2		-36.0	**	-32.4	**	28.1	**	35.4	**	28.8	**	33.2	*
L32/CML144	130.9	**	-12.5	**	9.5		48.7	**	49.0	**	1.5	-76.6	-13.9		-33.7	**	-22.5	**	30.0	**	29.0	**	14.0	**	5.4	
L33/CML144	96.9	**	-12.9	**	-18.1		39.4	**	23.6		-9.6	32.5	-3.0		-9.7		-20.7	**	21.2	**	33.6	**	10.2	*	18.5	
L34/CML144	98.9	**	-11.7	**	-159.7		43.0	**	50.1	**	4.8	-60.4	-3.2		-10.9		-17.4	*	26.6	**	25.5	**	3.6		1.2	
L35/CML144	129.0	**	-12.4	**	98.9		40.9	**	55.7	**	3.4	-13.1	19.4		-26.5	*	-21.8	**	36.8	**	31.6	**	13.5	**	2.1	
L36/CML144	101.3	**	-13.7	**	-92.6		44.7	**	50.3	**	1.7	38.3	11.0		-25.1	*	-9.7		28.3	**	28.2	**	10.1	*	0.8	
L37/CML144	117.4	**	-12.2	**	-498.7		48.5	**	47.5	**	-1.6	25.9	5.0		-26.2	*	-24.0	**	28.7	**	28.0	**	11.1	*	17.0	
L38/CML144	40.2		-13.5	**	-53.5		38.7	**	42.2	**	1.6	-2.4	-13.8		-17.6		-8.9		19.8	**	19.6	**	14.9	**	-13.6	
L39/CML144	90.0	**	-14.6	**	-214.6	*	38.2	**	47.5	**	8.4	-78.9	-16.0	*	-12.5		-33.5	**	22.7	**	18.0	*	10.1	*	14.1	
L40/CML144	94.7	**	-13.1	**	12.3		36.3	**	45.5	**	6.4	-23.7	-7.7		-16.0		-15.6	*	23.4	**	25.2	**	10.2	*	3.2	
L41/CML144	78.4	*	-11.7	**	-182.7		41.4	**	46.7	**	2.9	299.5	-1.0		-23.9	*	-28.0	**	18.3	*	15.0		14.1	**	1.8	
L42/CML144	97.8	**	-10.4	**	-140.1	*	46.3	**	30.5	*	-9.9	-88.8	-9.9		-25.3	*	-24.0	**	27.9	**	31.1	**	9.5	*	-15.2	
L43/CML144	75.3	*	-12.1	**	-44.8		39.0	**	27.6		-8.3	-27.8	-13.3		-26.6	*	-19.8	**	22.5	**	30.2	**	10.9	*	2.1	
L44/CML144	72.7	*	-11.5	**	-73.8		41.9	**	24.3		-12.6	-96.8	-9.0		-31.8	**	-17.0	*	23.3	**	31.8	**	6.4		28.5	
L45/CML144	88.9	**	-14.9	**	44.5		36.7	**	27.5		-7.2	-61.7	-19.1	*	-37.1	**	-25.9	**	29.7	**	37.8	**	6.4		26.1	
L46/CML144	52.7		-10.4	**	69.9		48.6	**	45.9	**	-1.0	-87.4	5.9		-9.8		-8.8		19.2	**	26.1	**	0.8		0.0	
L47/CML144	38.8		-11.4	**	52.4		46.1	**	31.4	*	-11.3	-12.4	-11.6		-36.8	**	-8.7		13.5	*	30.2	**	8.8	*	-9.8	
L48/CML144	63.9	*	-12.5	**	-284.9		46.3	**	38.5	**	-3.4	115.3	-4.3		-16.3		-18.7	*	23.3	**	30.4	**	5.4		-4.1	
L49/CML144	86.2	**	-12.7	**	-27.5		32.3	**	40.9	**	2.9	-36.9	7.6		-4.7		-20.1	**	18.9	**	16.3	*	6.8		-2.6	
L50/CML144	92.6	**	-12.2	**	-46.7		40.4	**	44.3	**	-3.6	-72.1	-4.0		-22.7	*	-24.9	**	30.6	**	32.3	**	8.3		8.5	
L51/CML144	73.5	*	-15.0	**	-82.9		40.1	**	43.0	**	-5.3	-52.9	-1.0		-11.0		-23.0	**	22.2	**	19.1	*	7.1		10.7	
L52/CML144	15.3		-14.4	**	-394.4		40.5	**	41.8	**	4.6	111.4	4.5		-12.5		-8.6		13.2	*	25.9	**	7.2		-8.8	
L53/CML144	126.4	**	-14.6	**	-48.3		41.9	**	58.4	**	12.3	37.4	-12.2		-16.2		-33.8	**	34.7	**	30.4	**	13.6	**	7.6	
L54/CML144	101.6	**	-13.2	**	-65.8		41.1	**	42.6	**	1.8	2.1	-9.4		-25.6	*	-34.1	**	26.3	**	22.4	**	8.7		9.5	
L55/CML144	104.8	**	-13.0	**	-115.3		37.5	**	36.0	*	-13.7	-59.8	-9.8		-21.7		-19.9	**	25.1	**	21.4	**	11.8	*	20.4	
L56/CML144	89.9	**	-13.3	**	-122.1		30.1	**	27.6		0.9	-98.2	-22.8	**	-22.3	*	-27.6	**	17.5	*	21.1	**	10.5	*	-0.9	
L57/CML144	88.7	**	-13.2	**	-103.8		37.3	**	41.2	**	-5.7	-40.2	-5.0		-11.3		-26.4	**	25.7	**	32.5	**	5.8		10.9	
L58/CML144	110.4	**	-14.7	**	-93.8		38.7	**	38.6	**	1.7	-84.7	-2.9		-19.9		-17.2	*	23.5	**	23.7	**	6.1		14.6	
L1/CML159	264.4	**	-12.3	**	2.6		59.8	**	86.6	**	3.4	-101.7	-9.3		-23.4	*	-23.1	**	17.3	*	55.9	**	9.1		9.9	
L2/CML159	294.0	**	-11.3	**	-8.1		42.2	**	50.7	**	0.4	229.0	-22.6	**	-37.7	**	-35.1	**	18.2	**	58.5	**	9.3		23.6	*

Appendix 5. Percentage better parent heterosis of QPM hybrids continued

Hybrids	GY		DA		ASI		PH		EH		EPO	SL	CLR		EA		PA		EL		KPR		RPE		TKW	
L3/CML159	262.3	**	-12.1	**	-30.0		47.4	**	43.6	**	-3.1	-81.9	-1.6		-27.2	**	-34.6	**	21.0	**	47.1	**	7.7		17.5	
L4/CML159	269.0	**	-11.5	**	23.5		40.5	**	44.8	**	3.9	115.0	-8.2		-27.9	**	-32.4	**	17.6	*	68.0	**	10.4	*	11.1	
L5/CML159	244.9	**	-11.0	**	-5.0		64.0	**	85.4	**	7.1	15.5	-16.6	*	-41.5	**	-21.0	**	18.3	**	51.5	**	6.8		27.4	*
L6/CML159	114.4	**	-11.3	**	-1.4		54.9	**	62.6	**	2.3	-109.9	-12.7		-24.1	**	-24.0	**	11.6		54.7	**	4.4		13.6	
L7/CML159	166.6	**	-12.3	**	96.1	*	52.5	**	58.6	**	-2.7	31.9	-15.0	*	-28.4	**	-20.5	**	14.8	*	48.3	**	6.7		21.0	
L8/CML159	211.0	**	-10.9	**	-26.8		59.2	**	77.5	**	1.0	23.6	-9.4		-28.0	**	-25.0	**	13.2		46.8	**	4.8		15.6	
L9/CML159	110.4	**	-11.6	**	27.4		54.2	**	75.5	**	3.1	216.3	-18.7	**	-34.7	**	-21.1	**	13.6		52.2	**	12.0	*	12.7	
L10/CML159	268.2	**	-13.8	**	12.8		56.8	**	79.3	**	4.3	-104.1	-5.0		-40.0	**	-32.3	**	12.0		45.6	**	10.7	*	28.1	*
L11/CML159	177.3	**	-11.2	**	-32.6		64.9	**	84.8	**	0.3	53.9	-5.8		-35.6	**	-25.6	**	14.8	*	51.2	**	4.5		5.7	
L12/CML159	141.8	**	-12.0	**	18.0		58.4	**	70.9	**	0.0	327.8	-16.1	*	-29.4	**	-28.7	**	15.0	*	54.9	**	7.5		14.2	
L13/CML159	143.3	**	-8.8	**	-35.0		55.8	**	68.4	**	5.1	58.0	-7.2		-32.7	**	-36.8	**	14.4	*	55.6	**	11.3	*	9.6	
L14/CML159	193.9	**	-9.6	**	-15.5		52.7	**	65.0	**	0.7	-35.5	-4.0		-40.6	**	-28.7	**	16.7	*	51.9	**	4.0		21.3	
L15/CML159	236.3	**	-9.1	**	68.0		56.7	**	48.4	**	-2.7	-95.2	-19.6	**	-33.6	**	-28.5	**	13.4		54.5	**	12.4	*	9.6	
L16/CML159	258.9	**	-12.3	**	-31.9		59.1	**	52.5	**	-3.5	50.3	-14.5	*	-39.8	**	-24.2	**	14.4	*	50.3	**	9.2		21.7	
L17/CML159	48.5		-12.1	**	33.5		33.2	**	33.6	*	-7.3	-58.1	-7.6		-27.2	**	-32.2	**	22.2	**	18.6	**	3.9		2.8	
L18/CML159	270.8	**	-11.7	**	-24.1		65.3	**	82.9	**	13.7	-81.5	1.0		-48.1	**	-27.9	**	23.7	**	54.8	**	8.1		12.4	
L19/CML159	256.9	**	-9.8	**	-11.2		56.5	**	39.4	**	-3.6	-66.6	-17.5	*	-30.2	**	-20.2	**	34.4	**	55.4	**	12.1	*	19.6	
L20/CML159	248.4	**	-12.3	**	-17.7		59.0	**	87.4	**	9.3	-36.4	-10.7		-45.0	**	-20.1	**	24.3	**	58.6	**	6.0		23.3	*
L21/CML159	255.0	**	-11.9	**	-50.6		53.0	**	59.0	**	0.5	-54.0	-8.0		-32.4	**	-27.1	**	31.8	**	60.4	**	12.1	*	14.6	
L22/CML159	167.5	**	-15.4	**	49.1		50.4	**	33.4	*	-6.9	-7.4	7.7		-33.6	**	-22.4	**	21.8	**	42.0	**	9.1		13.0	
L23/CML159	258.8	**	-13.8	**	40.3		60.6	**	62.5	**	9.1	-24.3	-14.1		-20.8	*	-34.8	**	18.0	**	51.6	**	3.3		17.1	
L24/CML159	274.5	**	-13.0	**	-34.4		63.5	**	83.2	**	12.2	-59.8	*	-12.5	-28.0	**	-32.2	**	14.4	*	60.6	**	10.8	*	8.3	
L25/CML159	140.9	**	-10.0	**	-27.2		48.1	**	57.3	**	6.9	40.6	-8.9		-30.6	**	-20.3	**	22.3	**	41.5	**	12.0	*	2.8	
L26/CML159	273.6	**	-11.8	**	-28.1		67.6	**	52.8	**	-3.5	-60.9	*	-20.5	**	-24.3	**	-30.4	**	14.2	*	57.6	**	3.3		11.5	
L27/CML159	230.2	**	-13.2	**	-7.2		66.0	**	77.5	**	2.5	-49.8	*	-10.2	-20.4	*	-21.8	**	11.6		39.9	**	8.5		9.7	
L28/CML159	269.4	**	-12.5	**	-106.3		66.8	**	72.0	**	2.3	-52.8	-23.5	**	-23.3	*	-29.7	**	25.5	**	53.8	**	5.1		17.1	
L29/CML159	225.4	**	-12.4	**	-26.9		62.5	**	63.1	**	5.6	-17.5	-9.7		-15.7		-18.5	**	15.6	*	44.2	**	7.5		6.6	
L30/CML159	312.2	**	-12.4	**	-29.0		61.7	**	63.5	**	-4.6	-70.3	-8.8		-42.8	**	-34.0	**	16.6	*	52.2	**	7.9		18.3	
L31/CML159	326.2	**	-12.7	**	-34.9		66.0	**	53.7	**	-12.2	-76.3	-17.8	*	-33.1	**	-37.5	**	12.6		57.9	**	10.8	*	19.7	
L32/CML159	217.4	**	-12.8	**	-72.2		64.4	**	97.3	**	11.9	-59.5	-18.6	*	-37.3	**	-26.5	**	20.4	**	59.0	**	4.7		14.8	

Appendix 5. Percentage better parent heterosis of QPM hybrids continued

Hybrids	GY		DA		ASI		PH		EH		EPO	SL	CLR		EA		PA		EL		KPR		RPE		TKW	
L33/CML159	120.9	**	-11.1	**	-50.2		41.9	**	35.2	*	-4.9	63.6	-4.6		-28.4	**	-15.7	*	15.2	*	43.3	**	9.8	*	13.1	
L34/CML159	275.6	**	-11.4	**	-123.1		53.4	**	85.3	**	13.5	-64.1	2.0		-31.2	**	-15.5	*	32.8	**	64.4	**	-0.7		29.4	*
L35/CML159	196.8	**	-10.9	**	-73.0		36.7	**	70.3	**	14.0	33.8	-0.9		-29.4	**	-29.9	**	31.3	**	49.4	**	7.3		28.1	*
L36/CML159	142.8	**	-12.5	**	-186.8		41.5	**	83.5	**	14.9	27.9	-0.3		-36.4	**	-19.1	*	26.0	**	41.1	**	4.0		31.0	**
L37/CML159	226.6	**	-10.2	**	-89.5		50.4	**	79.4	**	15.4	6.4	5.4		-42.3	**	-25.4	**	26.2	**	45.5	**	4.5		23.4	*
L38/CML159	54.8	*	-15.6	**	-27.2		33.6	**	51.2	*	-10.3	139.7	-18.4	**	-24.8	**	-16.0	*	21.1	**	26.4	**	8.9		-3.6	
L39/CML159	327.6	**	-14.1	**	-54.5		65.9	**	91.2	**	11.0	-22.7	-19.7	**	-35.7	**	-21.7	**	21.9	**	65.7	**	4.8		30.5	*
L40/CML159	119.5	**	-12.3	**	-105.9		56.5	**	64.3	**	8.7	-80.8	-7.0		-33.5	**	-13.6		21.3	**	31.9	**	0.1		16.3	
L41/CML159	280.5	**	-11.5	**	180.2		63.5	**	67.9	**	4.2	277.6	-6.3		-43.7	**	-28.9	**	18.2	**	52.9	**	14.8	**	21.5	
L42/CML159	241.3	**	-7.7	**	-35.9		59.5	**	71.0	**	2.3	-83.5	-10.4		-37.2	**	-20.6	**	20.4	**	55.2	**	2.7		4.3	
L43/CML159	282.1	**	-9.1	**	-46.3		56.8	**	82.5	**	9.2	-43.4	-13.5		-37.4	**	-15.7	*	26.7	**	43.0	**	10.7	*	11.4	
L44/CML159	223.8	**	-9.9	**	-64.1		61.0	**	73.8	**	2.9	-49.5	-8.7		-44.0	**	-20.8	**	24.0	**	42.1	**	0.7		1.1	
L45/CML159	274.6	**	-12.3	**	-6.9		64.9	**	55.7	**	-9.1	-28.0	-15.2		-45.2	**	-25.2	**	20.8	**	38.5	**	7.7		13.0	
L46/CML159	303.1	**	-8.2	**	35.7		66.2	**	69.9	**	-6.9	208.9	5.2		-37.2	**	-17.1	*	18.6	**	65.5	**	3.1		13.8	
L47/CML159	50.5	*	-10.6	**	149.0		37.6	**	60.8	**	6.7	36.0	-2.5		-37.7	**	-10.8		14.3	*	24.8	**	-4.8		6.8	
L48/CML159	90.2	**	-9.6	**	45.0		46.7	**	44.9	*	-5.6	675.8	-0.1		-31.5	**	-15.9	*	19.3	**	24.3	**	10.0	*	4.3	
L49/CML159	135.1	**	-11.0	**	-87.4		67.1	**	57.8	**	-7.3	-70.4	12.7		-33.9	**	-20.7	**	16.5	*	44.4	**	15.8	**	16.8	
L50/CML159	239.1	**	-12.4	**	-121.1		56.6	**	65.3	**	-11.6	-36.0	1.3		-31.4	**	-29.0	**	26.1	**	55.8	**	5.3		25.4	*
L51/CML159	279.3	**	-12.5	**	-34.6		61.8	**	57.9	**	-8.2	-52.8	-15.4		-40.6	**	-21.8	**	21.4	**	61.5	**	5.2		24.6	*
L52/CML159	70.7	**	-13.0	**	-138.6		49.6	**	37.3	*	-6.0	-77.1	3.1		-40.2	**	-15.1	*	16.8	**	24.6	**	11.1	*	8.4	
L53/CML159	220.5	**	-13.0	**	62.4		68.8	**	95.1	**	11.1	247.6	-12.1		-36.0	**	-31.0	**	22.2	**	42.5	**	6.3		27.0	*
L54/CML159	284.2	**	-13.3	**	-123.0		61.2	**	69.1	**	-3.1	174.7	5.7		-37.7	**	-27.2	**	20.8	**	52.0	**	6.2		30.9	**
L55/CML159	161.0	**	-10.7	**	-73.0		50.1	**	36.5	*	-10.7	-111.5	-4.0		-30.6	**	-20.4	**	17.7	*	53.5	**	3.6		24.7	*
L56/CML159	192.2	**	-13.0	**	-141.1		59.6	**	74.6	**	-2.6	-69.1	-28.1	**	-28.4	**	-20.1	**	16.9	*	55.0	**	4.7		14.2	
L57/CML159	201.5	**	-12.2	**	-73.9		57.4	**	60.0	**	-7.4	17.5	-5.2		-42.9	**	-20.1	**	23.1	**	55.9	**	3.8		32.9	**
L58/CML159	211.3	**	-12.5	**	-108.5	*	51.4	**	80.4	**	10.8	-95.0	8.3		-45.7	**	-16.8	*	17.2	*	43.9	**	0.7		29.9	*

Appendix 6. Percentage standard heterosis of QPM hybrids over MH140 evaluated at Melkassa, Edo Gojola and Mieso during 2014 main season

Hybrids	GY		DA		ASI	PH	EH	EPO		SL		CLR		EA	PA		EL		KPR	RPE	TKW	
L1/CML144	-17.2		-0.1		-44.5	2.1	-9.7	-9.8		20.2		71.3	**	6.6	23.9	*	-5.8		-0.4	5.8	-23.6	**
L2/CML144	0.9		-1.2		-86.1	8.3	-8.7	-12.7		45.0		46.2	**	-3.9	8.0		-2.8		8.0	5.1	-20.1	*
L3/CML144	-0.2		-1.2		-64.9	7.1	-14.2	-17.7	*	36.8		46.5	**	-5.8	1.0		-3.0		5.0	4.0	-13.6	
L4/CML144	3.9		0.6		-118.9	4.0	-4.0	3.3		94.1		45.5	**	-2.3	6.2		0.0		8.8	-1.1	-14.2	
L5/CML144	-18.2		0.9		2.5	-1.1	-10.5	-7.1		31.2		54.2	**	-1.8	6.0		-6.1		1.1	-1.1	-13.4	
L6/CML144	-16.0		-1.0		74.1	-1.9	-9.0	-5.0		-69.6		53.5	**	3.2	8.1		-6.9		-0.2	-2.6	-14.4	
L7/CML144	-17.3		-2.6		7.3	-1.7	-11.1	-7.4		-1.6		69.4	**	-3.6	11.4		-4.0		2.1	0.5	-13.7	
L8/CML144	-20.6		-1.3		18.6	-3.7	-14.6	-7.5		-15.0		55.3	**	5.4	12.4		-6.9		-2.2	-2.7	-18.4	*
L9/CML144	-28.4	*	-1.5		23.2	-3.1	-16.0	-11.9		63.4		61.0	**	8.4	16.7		-6.1		2.7	-3.6	-11.0	
L10/CML144	-27.7		-2.3		55.6	-2.8	-16.5	-13.3		233.0		54.6	**	3.2	12.1		-11.5	*	-5.3	3.8	-13.9	
L11/CML144	-21.2		-2.3		17.4	-1.6	-11.2	-4.9		-62.3		40.1	**	6.4	12.1		-6.5		-1.2	-0.7	-18.2	*
L12/CML144	-14.9		-2.8		8.9	-7.7	-16.6	-10.1		7.0		69.1	**	12.3	10.6		-9.7		-4.0	0.1	-15.9	
L13/CML144	-21.1		0.2		73.4	0.8	-4.6	-2.5		75.4		46.9	**	-7.6	18.8		-5.1		2.6	0.8	-25.8	**
L14/CML144	-15.4		-0.3		-12.7	2.1	-3.5	-2.9		286.3		62.2	**	-3.6	2.4		-5.6		6.2	-0.9	-27.3	**
L15/CML144	-14.5		1.0		-66.8	1.8	-8.3	-8.0		88.7		70.5	**	2.2	12.3		-8.0		3.1	5.2	-19.1	*
L16/CML144	-15.7		-0.4		-15.0	5.4	-3.3	-5.7		-14.9		55.8	**	1.5	0.1		-7.6		1.9	-0.5	-28.4	**
L17/CML144	-32.0	*	-3.5	*	42.4	-7.2	-14.2	-5.1		274.0		54.2	**	1.5	17.7		-5.1		-0.5	-1.2	-14.5	
L18/CML144	-18.5		-0.8		-53.1	3.7	5.6	6.7		505.1		54.4	**	6.7	27.0	**	5.7		9.2	0.7	-20.0	*
L19/CML144	-12.2		0.2		-56.8	7.1	4.2	0.5		252.4		69.8	**	-3.0	18.0		6.1		3.8	-2.8	-2.1	
L20/CML144	-15.2		-0.7		-58.3	6.9	0.5	-4.6		537.9		62.2	**	8.4	19.2		1.1		3.5	-1.6	-21.4	*
L21/CML144	-14.2		-1.5		-67.7	4.7	8.2	5.3		355.1		68.9	**	-6.4	11.8		0.8		3.3	1.6	-17.0	
L22/CML144	-20.2		-1.6		-0.9	-3.3	-6.4	-2.0		272.7		45.5	**	1.7	12.6		-1.9		11.8	1.7	-17.8	
L23/CML144	-14.1		-1.4		-83.1	3.2	5.7	8.8		1270.5	**	54.6	**	13.9	14.7		-2.0		10.7	0.0	-25.3	**
L24/CML144	-23.8		-2.5		-25.0	-0.3	2.4	8.9		1158.3	**	53.9	**	8.4	19.8		-7.1		1.5	0.4	-24.0	**
L25/CML144	-19.4		0.6		-48.8	3.3	2.5	3.2		761.7	*	55.5	**	11.7	18.4		-2.5		7.4	6.6	-27.0	**
L26/CML144	-28.2		-0.2		26.0	-0.5	-4.5	1.4		829.2	**	54.5	**	12.4	23.9	*	-11.9	*	2.6	1.7	-26.0	**
L27/CML144	-19.2		-2.3		-36.5	-0.7	2.3	8.1		1106.3	**	70.0	**	16.0	17.9		-4.0		3.0	2.4	-27.6	**
L28/CML144	-7.8		-1.8		-79.3	4.3	3.6	4.9		1062.3	**	70.0	**	16.0	17.3		0.0		2.6	3.0	-17.8	
L29/CML144	-28.0		0.9		-23.3	-0.7	1.1	4.9		964.1	**	61.4	**	26.5	12.2		-3.6		6.4	-3.1	-31.2	**
L30/CML144	-4.7		0.6		-107.0	5.5	-7.5	-9.3		-47.9		55.5	**	-19.4	6.9		-3.6		7.7	1.8	-16.0	

Appendix 6. Percentage standard heterosis of QPM hybrids over MH140 continued

Hybrids	GY	DA	ASI	PH	EH	EPO	SL	CLR	EA	PA	EL	KPR	RPE	TKW
L31/CML144	-4.0	-2.8	-72.8	0.0	-11.6	-9.3	-24.9	48.5 **	-14.2	22.6 *	-1.5	14.2 *	13.2 **	-11.0
L32/CML144	3.7	-0.6	-167.6 *	4.9	-3.5	-5.3	22.2	47.2 **	-19.0	23.6 *	0.0	8.8	0.2	-13.3
L33/CML144	-11.6	-1.0	-18.7	-1.7	-16.7	-12.1	142.9	32.6 *	10.4	11.8	-0.5	12.7	-2.4	-22.2 *
L34/CML144	-10.7	0.3	-63.1	0.8	-2.8	-2.2	-27.3	47.3 **	9.0	12.8	0.7	5.8	-9.0 *	-21.0 *
L35/CML144	2.8	-0.5	-160.0 *	6.9	0.9	-3.5	59.3	54.2 **	-10.1	1.6	5.2	10.9	-0.2	-19.7 *
L36/CML144	-9.6	-2.0	-104.6	6.7	-2.7	-5.2	153.5	54.6 **	-8.4	16.8	2.2	8.1	-3.2	-16.4
L37/CML144	-2.4	-0.3	-75.8	7.6	-4.5	-8.2	130.7	46.9 **	-9.8	9.1	-1.0	7.9	-2.3	-16.8
L38/CML144	-8.3	-1.8	-41.4	0.3	-7.9	-5.2	79.0	64.0 **	0.7	17.8	-3.1	5.3	8.5	-20.9 *
L39/CML144	-14.7	-3.0	-29.3	-2.6	-4.5	1.2	-61.4	53.5 **	7.0	5.7	-5.7	-0.5	-0.7	-23.2 *
L40/CML144	-10.7	-1.3	-143.6 *	-3.9	-5.8	-0.4	82.9	38.5 **	2.7	17.7	-5.1	5.5	-3.2	-14.9
L41/CML144	-19.9	0.3	-49.0	-0.3	-5.0	-2.6	632.4 *	55.0 **	2.2	1.4	-9.1	-3.0	0.3	-24.3 **
L42/CML144	-11.2	1.8	-145.1 *	3.2	-15.5	-15.9	-51.9	55.2 **	-8.6	13.3	-1.6	10.5	4.4	-28.7 **
L43/CML144	-21.3	-0.1	9.3	-2.0	-17.4	-14.5	113.4	47.1 **	-3.5	17.9	-5.8	9.8	1.4	-33.9 **
L44/CML144	-22.4	0.5	-54.1	0.1	-19.5 *	-18.5 *	-86.7	54.2 **	-4.7	18.2	-5.2	11.1	6.1	-29.8 **
L45/CML144	-15.2	-3.3 *	55.6	-3.6	-17.4	-13.5	-21.7	38.9 **	-11.3	11.3	-0.3	16.2 *	4.2	-19.1 *
L46/CML144	-31.5 *	1.8	105.6	4.8	-5.5	-7.6	-76.9	47.0 **	10.3	24.9 *	-8.3	6.3	0.2	-29.9 **
L47/CML144	-11.1	0.7	-5.7	5.7	-14.9	-17.3 *	120.1	40.2 **	-22.7	18.1	-6.8	10.4	13.7 **	-22.2 *
L48/CML144	-15.1	-0.7	-29.4	3.2	-10.3	-9.9	294.8	47.1 **	2.4	6.5	-5.2	10.0	1.1	-25.2 **
L49/CML144	-16.4	-0.9	-144.8 *	-6.7	-8.7	1.3	15.8	46.5 **	16.5	19.1	-8.5	-1.9	-6.1	-21.8 *
L50/CML144	-13.5	-0.3	-132.9 *	-1.0	-6.5	-1.6	-33.3	46.4 **	-5.5	18.5	0.4	11.5	-4.8	-19.5 *
L51/CML144	-22.1	-3.4 *	-110.6	-1.2	-7.4	-5.1	-13.7	54.1 **	10.7	17.8	-6.1	0.4	-5.9	-20.2 *
L52/CML144	-19.2	-2.7	-32.5	-0.9	-8.2	-2.4	287.6	47.5 **	7.0	18.2	1.2	10.6	-4.1	-17.4
L53/CML144	1.7	-2.9	-61.4	0.1	2.6	4.8	152.0	54.1 **	2.5	12.9	3.6	9.9	-0.1	-16.2
L54/CML144	-9.5	-1.4	-121.1	-0.5	-7.7	-4.2	87.2	39.0 **	-0.1	7.0	-2.9	3.2	-4.1	-18.6 *
L55/CML144	-8.0	-1.2	-90.6	-3.0	-11.9	-5.6	-26.4	38.0 **	-4.3	19.0	-3.8	2.3	-1.8	-16.4
L56/CML144	-14.8	-1.5	-86.3	-8.3	-17.4	-4.7	-95.6	40.2 **	9.1	6.7	-9.6	2.1	-2.9	-18.7 *
L57/CML144	-15.3	-1.4	-97.7	-3.2	-8.5	-1.8	9.7	47.3 **	8.5	1.7	-3.4	11.7	-7.0	-14.0
L58/CML144	-5.6	-3.1	-92.4	-2.2	-10.3	-5.1	-21.7	38.1 **	-2.1	18.8	-5.0	4.3	-4.7	-16.1
L1/CML159	-13.1	-3.5 *	45.1	4.8	-13.2	-15.4	-101.1	54.9 **	15.8	10.4	-5.6	3.5	0.1	-15.5
L2/CML159	-6.0	-2.4	-43.3	-0.4	-18.7 *	-17.9 *	237.0	54.6 **	-5.3	2.4	-4.9	5.2	0.9	-5.0

Appendix 6. Percentage standard heterosis of QPM hybrids over MH140 continued

Hybrids	GY	DA		ASI		PH	EH		EPO		SL		CLR		EA	PA		EL	KPR	RPE	TKW	
L3/CML159	-1.8	-3.2	*	16.7		7.7	-19.6	*	-20.7	*	-85.1		63.5	**	10.1	0.4		-2.6	4.1	-1.2	-9.7	
L4/CML159	-1.7	-2.6		-2.2		1.3	-15.5		-15.0		35.7		54.0	**	9.0	-5.3		-5.4	12.3	1.3	-14.6	
L5/CML159	2.8	-2.1		28.5		0.7	-13.8		-12.4		5.4		55.4	**	-11.5	10.8		-4.8	0.5	-2.0	0.6	
L6/CML159	-20.2	-2.4		101.4		-1.7	-20.5	*	-16.3		-106.2		61.8	**	14.8	6.5		-10.3	2.7	-4.2	-7.6	
L7/CML159	-15.4	-3.4	*	182.8	**	-6.3	-26.2	**	-19.4	*	-16.8		55.0	**	8.3	11.5		-7.7	-1.6	-2.0	-7.0	
L8/CML159	-9.6	-2.0		19.5		-2.2	-16.1		-12.0		-22.0		54.2	**	8.9	5.2		-8.9	-2.6	-3.8	-5.6	
L9/CML159	-20.4	-2.7		74.6		-5.3	-18.4	*	-12.0		99.5		62.4	**	-1.3	10.7		-8.6	1.0	2.8	-12.7	
L10/CML159	-12.2	-5.1	**	156.1	*	-3.7	-16.6		-11.2		-102.6		78.1	**	-9.2	8.8		-9.9	-3.4	1.6	-1.5	
L11/CML159	-13.0	-2.3		48.9		1.3	-13.8		-11.7		-2.9		62.0	**	-2.5	6.4		-7.7	0.3	-4.1	-12.5	
L12/CML159	-9.8	-3.1		83.6		-2.7	-20.6	*	-18.2	*	169.9		54.0	**	6.8	0.0		-7.5	2.8	3.1	-12.2	
L13/CML159	-7.3	0.3		-0.2		3.9	-12.4		-12.5		85.5		45.5	**	1.8	-11.4		-8.0	3.8	2.1	-15.5	
L14/CML159	-3.5	-0.5		20.6		1.2	-10.7		-12.5		-29.8		61.3	**	-10.2	-0.1		-6.1	2.4	-4.6	-6.7	
L15/CML159	-4.0	0.1		113.5		2.3	-11.6		-9.8		-89.8		46.8	**	0.4	0.2		-8.8	2.5	3.1	-15.7	
L16/CML159	-5.1	-3.4	*	17.0		3.0	-16.0		-16.8		85.8		69.5	**	-8.9	6.3		-7.9	-0.3	0.2	-6.4	
L17/CML159	-19.8	-3.3	*	19.6		-0.8	-16.9		-14.8		11.5		38.7	**	10.1	-4.9		-1.7	8.2	-4.7	-3.0	
L18/CML159	12.7	-2.8		-22.7		6.4	-5.3		-7.0		37.6		55.8	**	-21.5	4.7		-0.5	5.3	-0.8	-13.6	
L19/CML159	-1.2	-0.7		20.2		7.9	-7.2		-11.4		173.5		55.1	**	5.5	20.0		8.1	5.3	2.8	-8.0	
L20/CML159	0.1	-3.4	*	22.7		2.6	-8.7		-10.2		390.1		53.4	**	-16.8	12.0		0.0	5.3	-2.7	-5.2	
L21/CML159	2.2	-3.0		-67.2		8.1	-6.5		-10.8		309.8		54.6	**	2.3	9.1		6.0	6.4	2.9	-11.3	
L22/CML159	-6.7	-6.9	**	-8.0		1.5	-16.0		-14.4		-41.6		46.8	**	0.4	8.8		-2.0	0.5	3.5	-13.2	
L23/CML159	-10.4	-5.1	**	14.9		-0.7	-6.2		-0.4		567.1		46.5	**	19.8	7.8		-5.1	9.0	-3.4	-9.9	
L24/CML159	-10.6	-4.2	**	-22.7		0.7	2.4		3.5		426.5		62.6	**	8.9	17.3		-8.0	6.5	1.7	-16.7	
L25/CML159	-6.6	-1.0		-55.1		7.0	3.4		0.3		1070.3	**	53.4	**	5.0	11.8		-1.6	3.5	3.5	-21.0	*
L26/CML159	-10.9	-2.9		16.6		2.9	-6.7		-6.7		351.0		47.0	**	14.6	19.9		-8.1	4.5	0.6	-14.3	
L27/CML159	-9.6	-4.4	**	-42.7		2.0	-2.3		-0.8		570.0		54.5	**	20.3	12.8		-10.3	0.0	-0.4	-15.6	
L28/CML159	-3.4	-3.7	*	-103.9		3.8	-0.8		-1.3		424.5		54.0	**	16.0	17.8		0.9	7.0	-3.6	-10.0	
L29/CML159	-18.4	-3.6	*	-54.9		-0.2	1.6		4.5		871.2	**	53.7	**	27.5	24.0	*	-7.0	2.3	-1.3	-18.0	*
L30/CML159	-1.7	-3.6	*	-56.2		-0.7	-13.4		-10.3		-43.3		54.2	**	-10.6	7.8		-6.2	1.0	-0.9	-9.1	
L31/CML159	1.7	-3.9	*	-38.3		2.0	-14.6		-13.1		56.0		31.5	*	1.1	13.4		-9.4	7.2	1.7	-8.0	
L32/CML159	-1.5	-4.0	*	-82.8		1.0	-8.3		-7.4		111.4		39.2	**	-5.2	17.3		-3.1	5.5	-3.9	-5.6	

Appendix 6. Percentage standard heterosis of QPM hybrids over MH140 continued

Hybrids	GY	DA		ASI		PH	EH		EPO		SL	CLR		EA	PA		EL	KPR	RPE		TKW	
L33/CML159	-12.6	-2.1		-50.5		-1.0	-8.9		-7.5		180.8	30.4	*	8.3	18.9		-5.4	3.7	0.8		-13.0	
L34/CML159	7.4	-2.5		-114.2		4.7	-6.1		-7.1		-77.3	55.2	**	4.1	18.5		6.8	9.1	-8.8	*	1.0	
L35/CML159	11.6	-1.9		-83.3		3.7	-3.6		-6.7		34.0	22.7		6.8	-1.7		5.7	6.2	-1.5		0.7	
L36/CML159	-7.5	-3.6	*	-153.5	*	4.4	-2.9		-6.0		-19.3	38.8	**	-3.8	13.4		1.3	2.6	-4.5		8.5	
L37/CML159	8.7	-1.2		-93.5		9.0	-1.4		-5.6		-32.9	47.4	**	-12.7	7.1		1.5	3.2	-4.1		-5.1	
L38/CML159	1.3	-7.1	**	-8.2		-3.4	-29.7	**	-26.6	**	233.3	55.2	**	13.8	17.7		-2.1	11.4	2.9		-11.8	
L39/CML159	2.0	-5.4	**	-71.9		1.9	-11.1		-9.1		36.5	46.7	**	-2.8	24.5	*	-1.9	10.0	-3.8		0.3	
L40/CML159	0.7	-3.4	*	-103.6		0.8	-1.5		1.8		-54.1	39.6	**	0.6	21.1	*	-2.4	9.2	-8.1		-4.0	
L41/CML159	1.2	-2.6		72.9		3.6	-0.8		-1.4		138.3	46.8	**	-14.8	0.1		-4.9	1.5	5.3		-6.6	
L42/CML159	-18.6	1.6		-27.8		-2.0	-20.5	*	-16.3		-29.3	54.3	**	-5.0	18.4		-3.2	3.0	-2.0		-12.4	
L43/CML159	-8.8	0.1		6.3		-0.1	-14.1		-10.6		67.2	46.8	**	-5.3	23.9	*	1.9	7.8	1.6		-14.3	
L44/CML159	-7.6	-0.8		-37.2		0.8	-19.2	*	-15.8		110.1	54.8	**	-15.4	12.7		-0.2	9.2	0.5		-22.3	*
L45/CML159	8.5	-3.4	*	0.2		1.3	-27.6	**	-25.6	**	47.3	45.7	**	-17.1	12.2		-2.9	10.7	5.4		-13.1	
L46/CML159	-3.8	1.0		64.2		4.2	-21.0	*	-23.8	**	94.9	45.9	**	-5.1	16.2		-4.6	9.8	2.5		-12.5	
L47/CML159	-3.6	-1.6		54.1		-0.5	-15.8		-12.7		241.8	54.6	**	-5.8	25.0	*	-6.2	5.8	-0.6		-7.9	
L48/CML159	-1.5	-0.5		-10.5		-7.5	-25.8	**	-20.5	*	389.5	53.5	**	3.6	17.9		-4.1	3.1	5.5		-18.7	*
L49/CML159	-7.8	-2.0		-92.2		2.6	-10.1		-8.8		-66.4	53.5	**	-0.1	18.2		-6.3	1.5	6.2		-6.2	
L50/CML159	0.0	-3.6	*	-113.0		-3.8	-15.7		-9.8		53.2	54.5	**	3.8	12.1		1.4	6.4	-3.4		-3.6	
L51/CML159	-1.6	-3.7	*	-59.7		-0.7	-11.9		-8.0		-32.2	31.7	*	-10.1	19.5		-2.4	7.2	-3.4		-4.2	
L52/CML159	19.7	-4.3	**	-123.8		1.9	-17.7		-18.1	*	-72.4	45.5	**	-9.5	19.0		4.4	9.5	1.9		-1.8	
L53/CML159	7.9	-4.2	**	21.3		3.7	-9.3		-7.3		212.1	54.2	**	-3.2	17.7		-1.7	-5.5	-2.5		-1.1	
L54/CML159	1.2	-4.5	**	-114.2		-1.0	-12.3		-8.8		73.3	62.2	**	-5.8	18.2		-2.8	0.8	-2.5		0.7	
L55/CML159	-4.2	-1.7		-83.3		-7.8	-12.9		-2.3		-108.7	46.9	**	4.9	18.4		-5.3	1.8	-4.9		-4.1	
L56/CML159	-10.5	-4.3	**	-125.4		-2.0	-11.5		-8.0		-25.4	30.7	*	8.3	17.7		-5.9	2.9	-4.0		-6.3	
L57/CML159	8.1	-3.4	*	-83.9		-3.3	-10.3		-3.7		-25.9	47.0	**	-13.6	12.0		-1.0	5.0	-4.7		3.1	
L58/CML159	3.7	-3.6	*	-110.4		-2.3	-13.3		-9.3		-74.3	54.0	**	-17.8	19.3		-5.7	2.2	-7.6		-0.1	

Appendix 7. Percentage standard heterosis of QPM hybrids over MH130 evaluated at Melkassa, Edo Gojola and Mieso during 2014 main season

Hybrids	GY		DA		ASI	PH		EH		EPO		SL		CLR	EA		PA	EL	KPR	RPE	TKW	
L1/CML144	-16.1		10.5	**	-13.7	7.8		23.1		11.7		-52.7		10.5	19.7		5.2	-1.6	-3.1	4.5	-25.7	**
L2/CML144	2.4		9.3	**	-78.3	14.4	**	24.6		8.0		-42.9		-5.7	7.9		-8.3	1.5	5.1	3.8	-22.2	*
L3/CML144	1.2		9.4	**	-45.4	13.1	*	17.1		1.9		-46.1		-5.5	5.8		-14.2	1.3	2.1	2.8	-15.9	
L4/CML144	5.4		11.4	**	-129.3	9.8		30.9	*	27.8	**	-23.6		-6.1	9.6		-9.8	4.4	5.9	-2.3	-16.5	
L5/CML144	-17.1		11.7	**	59.4	4.4		22.1		15.0		-48.4		-0.5	10.2		-10.0	-1.9	-1.6	-2.2	-15.7	
L6/CML144	-14.8		9.5	**	170.8	3.6		24.2		17.6		-88.1		-1.0	15.8		-8.2	-2.8	-2.9	-3.8	-16.7	
L7/CML144	-16.1		7.7	**	66.9	3.9		21.2		14.5		-61.3		9.3	8.3		-5.4	0.2	-0.7	-0.7	-16.0	
L8/CML144	-19.4		9.3	**	84.4	1.6		16.4		14.4		-66.5		0.2	18.4		-4.5	-2.7	-4.9	-3.8	-20.6	*
L9/CML144	-27.4		9.0	**	91.6	2.3		14.6		9.0		-35.7		3.9	21.7		-0.9	-1.9	-0.1	-4.7	-13.4	
L10/CML144	-26.7		8.1	**	142.0	2.7		13.9		7.3		31.1		-0.2	15.8		-4.8	-7.6	-7.8	2.6	-16.2	
L11/CML144	-20.1		8.1	**	82.6	3.9		21.1		17.7		-85.2		-9.6	19.4		-4.8	-2.4	-3.9	-1.9	-20.4	*
L12/CML144	-13.7		7.6	**	69.4	-2.6		13.8		11.3		-57.9		9.1	26.0		-6.1	-5.7	-6.6	-1.1	-18.1	*
L13/CML144	-19.9		10.9	**	169.7	6.4		30.1	*	20.7		-31.0		-5.2	3.7		0.9	-0.9	-0.2	-0.4	-27.8	**
L14/CML144	-14.1		10.3	**	35.7	7.8		31.6	*	20.2		52.1		4.6	8.2		-13.0	-1.4	3.3	-2.1	-29.3	**
L15/CML144	-13.3		11.8	**	-48.3	7.5		25.0		13.9		-25.7		10.0	14.7		-4.6	-3.9	0.3	4.0	-21.3	*
L16/CML144	-14.5		10.2	**	32.2	11.3	*	31.9	*	16.7		-66.5		0.5	14.0		-15.0	-3.5	-0.8	-1.7	-30.3	**
L17/CML144	-31.0	*	6.7	**	121.4	-2.0		17.0		17.5		47.2		-0.5	13.9		0.0	-0.9	-3.3	-2.4	-16.8	
L18/CML144	-17.3		9.8	**	-27.0	9.5		44.0	**	32.0	**	138.2		-0.4	19.8		7.9	10.4	6.3	-0.5	-22.1	*
L19/CML144	-11.0		10.9	**	-32.8	13.1	*	42.2	**	24.4	*	38.7		9.5	8.9		0.3	10.8	1.0	-3.9	-4.7	
L20/CML144	-14.0		9.9	**	-35.2	12.9	*	37.0	**	18.0		151.1		4.7	21.7		1.3	5.6	0.7	-2.7	-23.5	**
L21/CML144	-13.0		9.0	**	-49.8	10.6	*	47.6	**	30.4	**	79.1		9.0	5.1		-5.1	5.2	0.5	0.4	-19.3	*
L22/CML144	-19.1		8.9	**	54.2	2.1		27.7	*	21.2	*	46.7		-6.1	14.1		-4.3	2.4	8.7	0.5	-20.0	*
L23/CML144	-12.8		9.1	**	-73.7	9.0		44.1	**	34.6	**	439.5	**	-0.3	27.9		-2.6	2.3	7.7	-1.2	-27.3	**
L24/CML144	-22.7		7.9	**	16.6	5.3		39.6	**	34.8	**	395.3	**	-0.7	21.7		1.8	-2.9	-1.3	-0.8	-26.0	**
L25/CML144	-18.3		11.3	**	-20.3	9.1		39.8	**	27.8	**	239.2		0.3	25.3		0.6	1.9	4.5	5.3	-29.0	**
L26/CML144	-27.1		10.4	**	95.9	5.1		30.3	*	25.4	*	265.8	*	-0.3	26.1		5.3	-8.0	-0.2	0.5	-28.0	**
L27/CML144	-18.1		8.2	**	-1.3	4.8		39.5	**	33.8	**	374.8	**	9.6	30.2		0.2	0.3	0.2	1.1	-29.5	**
L28/CML144	-6.5		8.6	**	-67.8	10.1		41.3	**	29.9	**	357.5	**	9.6	30.2		-0.4	4.5	-0.2	1.8	-20.0	*
L29/CML144	-27.0		11.6	**	19.3	4.9		37.9	**	29.8	**	318.9	**	4.1	42.0	**	-4.7	0.7	3.5	-4.3	-33.0	**
L30/CML144	-3.4		11.4	**	-110.8	11.4	*	26.1	*	12.3		-79.5		0.3	-9.5		-9.2	0.7	4.8	0.6	-18.3	*

Appendix 7. Percentage standard heterosis of QPM hybrids over MH130 continued

Hybrids	GY		DA		ASI		PH		EH		EPO		SL	CLR	EA	PA	EL	KPR	RPE		TKW	
L31/CML144	-2.6		7.5	**	-57.7		5.6		20.6		12.3		-70.4	-4.2	-3.6	4.2	2.9	11.1	11.9	**	-13.3	
L32/CML144	5.2		10.0	**	-205.1	*	10.7	*	31.6	*	17.2		-51.9	-5.0	-9.1	5.0	4.4	5.8	-1.0		-15.6	
L33/CML144	-10.3		9.5	**	26.5		3.8		13.6		8.8		-4.4	-14.5	23.9	-5.0	3.9	9.6	-3.5		-24.3	**
L34/CML144	-9.5		11.0	**	-42.7		6.5		32.6	*	21.0	*	-71.4	-4.9	22.3	-4.2	5.1	2.9	-10.0	*	-23.1	**
L35/CML144	4.3		10.1	**	-193.3		12.9	*	37.6	**	19.4		-37.3	-0.5	0.9	-13.7	9.9	7.9	-1.4		-21.8	*
L36/CML144	-8.3		8.4	**	-107.1		12.7	*	32.7	*	17.4		-0.2	-0.3	2.8	-0.8	6.8	5.2	-4.4		-18.7	*
L37/CML144	-1.0		10.4	**	-62.4		13.7	**	30.3	*	13.6		-9.2	-5.2	1.3	-7.3	3.3	4.9	-3.5		-19.1	*
L38/CML144	-7.0		8.7	**	-8.8		5.9		25.6	*	17.4		-29.5	5.8	13.0	0.1	1.2	2.5	7.2		-23.0	**
L39/CML144	-13.5		7.3	**	10.0		2.9		30.3	*	25.2	*	-84.8	-1.0	20.1	-10.3	-1.5	-3.2	-1.8		-25.2	**
L40/CML144	-9.4		9.3	**	-167.8		1.5		28.5	*	23.3	*	-28.0	-10.7	15.3	-0.1	-0.9	2.7	-4.3		-17.2	
L41/CML144	-18.8		11.0	**	-20.7		5.3		29.6	*	20.5		188.3	0.0	14.7	-13.9	-5.0	-5.7	-0.9		-26.3	**
L42/CML144	-9.9		12.6	**	-170.2		9.0		15.3		4.1		-81.1	0.1	2.6	-3.8	2.7	7.5	3.2		-30.6	**
L43/CML144	-20.2		10.5	**	69.9		3.5		12.7		5.9		-16.0	-5.1	8.4	0.2	-1.7	6.8	0.2		-35.7	**
L44/CML144	-21.3		11.2	**	-28.6		5.7		9.8		0.9		-94.8	-0.5	7.0	0.4	-1.0	8.1	4.8		-31.7	**
L45/CML144	-14.0		7.0	**	142.0		1.8		12.6		7.1		-69.2	-10.4	-0.4	-5.5	4.1	13.0	* 2.9		-21.2	*
L46/CML144	-30.5	*	12.6	**	219.8	*	10.7	*	28.9	*	14.3		-90.9	-5.2	23.9	6.1	-4.2	3.5	-1.0		-31.8	**
L47/CML144	-9.8		11.4	**	46.6		11.6	*	16.1		2.4		-13.4	-9.6	-13.2	0.3	-2.7	7.4	12.4	**	-24.3	**
L48/CML144	-13.9		9.9	**	9.8		8.9		22.4		11.5		55.4	-5.1	14.9	-9.5	-1.0	7.0	-0.1		-27.2	**
L49/CML144	-15.2		9.7	**	-169.6		-1.5		24.5		25.3	*	-54.4	-5.5	30.8	1.1	-4.5	-4.6	-7.2		-23.9	**
L50/CML144	-12.3		10.4	**	-151.2		4.5		27.5	*	21.8	*	-73.8	-5.5	6.0	0.6	4.9	8.5	-6.0		-21.7	*
L51/CML144	-21.0		6.9	**	-116.4		4.4		26.3	*	17.5		-66.0	-0.6	24.3	0.0	-1.9	-2.3	-7.0		-22.3	*
L52/CML144	-18.0		7.6	**	5.0		4.7		25.2	*	20.8		52.6	-4.8	20.1	0.4	5.7	7.6	-5.2		-19.6	*
L53/CML144	3.1		7.4	**	-39.9		5.7		39.9	**	29.7	**	-0.8	-0.6	15.0	-4.1	8.2	6.9	-1.3		-18.4	*
L54/CML144	-8.2		9.2	**	-132.8		5.1		25.9	*	18.6		-26.3	-10.3	12.2	-9.1	1.5	0.4	-5.2		-20.8	*
L55/CML144	-6.7		9.4	**	-85.3		2.4		20.1		16.9		-71.0	-11.0	7.5	1.1	0.4	-0.4	-2.9		-18.7	*
L56/CML144	-13.5		9.0	**	-78.8		-3.1		12.7		17.9		-98.3	-9.6	22.5	-9.4	-5.6	-0.7	-4.1		-20.8	*
L57/CML144	-14.1		9.1	**	-96.4		2.3		24.7		21.5	*	-56.8	-5.0	21.8	-13.6	0.9	8.7	-8.1		-16.3	
L58/CML144	-4.2		7.3	**	-88.2		3.3		22.4		17.4		-69.2	-10.9	9.9	0.9	-0.8	1.5	-5.9		-18.3	*
L1/CML159	-11.8		6.8	**	125.6		10.7	*	18.4		4.7		-100.4	-0.1	30.0	-6.3	-1.4	0.6	-1.1		-17.8	*
L2/CML159	-4.7		8.0	**	-11.8		5.2		10.8		1.6		32.6	-0.2	6.3	-13.0	-0.7	2.3	-0.3		-7.5	

Appendix 7. Percentage standard heterosis of QPM hybrids over MH130 continued

Hybrids	GY	DA		ASI		PH		EH		EPO		SL		CLR	EA		PA		EL		KPR	RPE	TKW	
L3/CML159	-0.4	7.1	**	81.5		13.7	**	9.6		-1.8		-94.1		5.5	23.5		-14.7		1.7		1.3	-2.4	-12.1	
L4/CML159	-0.3	7.7	**	52.0		6.9		15.3		5.2		-46.6		-0.7	22.3		-19.5	*	-1.2		9.3	0.1	-16.9	
L5/CML159	4.3	8.4	**	99.8		6.4		17.6		8.5		-58.5		0.2	-0.7		-5.9		-0.6		-2.2	-3.2	-2.1	
L6/CML159	-19.0	8.0	**	213.3	*	3.8		8.4		3.6		-102.4		4.4	28.9		-9.5		-6.3		-0.1	-5.3	-10.1	
L7/CML159	-14.2	6.9	**	339.8	**	-1.1		0.6		-0.3		-67.2		0.0	21.5		-5.3		-3.6		-4.3	-3.2	-9.5	
L8/CML159	-8.3	8.5	**	85.8		3.2		14.5		8.9		-69.3		-0.5	22.2		-10.7		-4.9		-5.2	-5.0	-8.1	
L9/CML159	-19.3	7.7	**	171.6		0.0		11.3		8.9		-21.5		4.8	10.8		-6.0		-4.6		-1.8	1.6	-15.0	
L10/CML159	-10.9	5.0	**	298.3	**	1.7		13.7		9.9		-101.0		14.9	1.9		-7.6		-5.9		-6.0	0.4	-4.2	
L11/CML159	-11.8	8.1	**	131.5		6.9		17.6		9.3		-61.8		4.5	9.4		-9.6		-3.6		-2.4	-5.3	-14.9	
L12/CML159	-8.5	7.2	**	185.5		2.7		8.4		1.2		6.2		-0.6	19.9		-15.1		-3.4		0.0	1.9	-14.6	
L13/CML159	-6.0	11.0	**	55.2		9.7		19.4		8.3		-27.0		-6.2	14.2		-24.8	**	-3.9		1.0	0.9	-17.8	*
L14/CML159	-2.1	10.1	**	87.6		6.8		21.8		8.3		-72.4		4.0	0.8		-15.1		-2.0		-0.4	-5.7	-9.2	
L15/CML159	-2.6	10.7	**	232.1	*	8.0		20.5		11.7		-96.0		-5.3	12.7		-14.9		-4.8		-0.3	1.9	-18.0	*
L16/CML159	-3.7	6.9	**	81.9		8.7		14.5		3.0		-26.9		9.3	2.3		-9.7		-3.9		-3.0	-1.0	-8.9	
L17/CML159	-18.7	7.0	**	85.9		4.7		13.3		5.5		-56.1		-10.6	23.6		-19.2	*	2.7		5.3	-5.8	-5.6	
L18/CML159	14.3	7.6	**	20.2		12.3	*	29.2	*	15.1		-45.8		0.5	-11.9		-11.1		3.9		2.4	-1.9	-15.9	
L19/CML159	0.2	9.9	**	86.9		13.9	**	26.6	*	9.7		7.7		0.1	18.4		1.9		12.9	*	2.4	1.6	-10.5	
L20/CML159	1.5	6.8	**	90.9		8.4		24.5		11.2		92.9		-1.0	-6.6		-4.9		4.4		2.4	-3.8	-7.7	
L21/CML159	3.7	7.3	**	-48.9		14.2	**	27.5	*	10.4		61.3		-0.3	14.8		-7.3		10.7		3.5	1.7	-13.6	
L22/CML159	-5.4	3.0		43.1		7.2		14.6		5.9		-77.0		-5.3	12.6		-7.6		2.3		-2.3	2.3	-15.5	
L23/CML159	-9.1	5.0	**	78.7		4.9		28.0	*	23.3	*	162.6		-5.5	34.5	*	-8.4		-0.8		6.0	-4.5	-12.4	
L24/CML159	-9.4	6.0	**	20.3		6.4		39.7	**	28.0	**	107.2		4.9	22.2		-0.4		-3.9		3.6	0.5	-19.0	*
L25/CML159	-5.3	9.6	**	-30.1		13.0	*	41.0	**	24.2	*	360.7	**	-1.0	17.9		-5.1		2.7		0.7	2.2	-23.1	**
L26/CML159	-9.6	7.5	**	81.4		8.7		27.2	*	15.5		77.5		-5.2	28.6		1.9		-4.1		1.7	-0.6	-16.6	
L27/CML159	-8.3	5.8	**	-11.0		7.7		33.3	**	22.8	*	163.7		-0.3	35.1	*	-4.2		-6.3		-2.7	-1.6	-17.9	*
L28/CML159	-2.0	6.6	**	-106.1		9.7		35.3	**	22.1	*	106.5		-0.7	30.2		0.0		5.4		4.0	-4.7	-12.4	
L29/CML159	-17.2	6.7	**	-29.8		5.4		38.6	**	29.3	**	282.3	*	-0.9	43.1	**	5.3		-2.9		-0.5	-2.5	-20.2	*
L30/CML159	-0.3	6.7	**	-31.8		4.9		18.1		11.1		-77.7		-0.6	0.4		-8.5		-2.0		-1.8	-2.1	-11.5	
L31/CML159	3.1	6.3	**	-4.1		7.7		16.5		7.6		-38.6		-15.2	13.5		-3.7		-5.4		4.3	0.5	-10.4	
L32/CML159	-0.1	6.2	**	-73.3		6.6		25.1	*	14.6		-16.8		-10.2	6.5		-0.4		1.2		2.6	-5.1	-8.1	

Appendix 7. Percentage standard heterosis of QPM hybrids over MH130 continued

Hybrids	GY	DA		ASI	PH		EH		EPO		SL	CLR		EA	PA	EL		KPR	RPE		TKW	
L33/CML159	-11.4	8.3	**	-23.1	4.6		24.3		14.5		10.5	-15.9		21.6	0.9	-1.2		0.8	-0.4		-15.4	
L34/CML159	8.9	7.9	**	-122.1	10.6	*	28.0	*	15.0		-91.1	0.1		16.9	0.6	11.5	*	6.1	-9.9	*	-1.7	
L35/CML159	13.2	8.5	**	-74.1	9.5		31.5	*	15.5		-47.2	-20.8	*	19.9	-16.5	10.3		3.3	-2.7		-2.0	
L36/CML159	-6.2	6.6	**	-183.3	10.2		32.5	*	16.3		-68.2	-10.4		8.0	-3.7	5.8		-0.2	-5.7		5.6	
L37/CML159	10.2	9.4	**	-89.9	15.1	**	34.5	**	16.9		-73.6	-4.9		-2.0	-9.1	6.1		0.4	-5.2		-7.7	
L38/CML159	2.7	2.8		42.7	2.0		-4.1		-9.1		31.2	0.1		27.7	0.0	2.3		8.3	1.6		-14.2	
L39/CML159	3.5	4.7	**	-56.3	7.6		21.2		12.5		-46.3	-5.4		9.1	5.8	2.4		7.0	-5.0		-2.3	
L40/CML159	2.1	6.8	**	-105.6	6.5		34.4	**	26.0	*	-81.9	-9.9		13.0	2.8	1.9		6.2	-9.2	*	-6.6	
L41/CML159	2.6	7.8	**	168.9	9.4		35.2	**	22.0	*	-6.2	-5.3		-4.4	-15.0	-0.7		-1.3	4.1		-9.1	
L42/CML159	-17.4	12.4	**	12.2	3.5		8.4		3.6		-72.2	-0.5		6.6	0.6	1.1		0.2	-3.2		-14.7	
L43/CML159	-7.5	10.7	**	65.3	5.5		17.2		10.6		-34.2	-5.3		6.3	5.3	6.4		4.9	0.4		-16.6	
L44/CML159	-6.3	9.7	**	-2.3	6.5		10.2		4.2		-17.3	-0.2		-5.0	-4.2	4.2		6.2	-0.7		-24.4	**
L45/CML159	10.1	6.8	**	55.8	6.9		-1.3		-7.9		-42.0	-6.0		-7.0	-4.7	1.4		7.6	4.1		-15.4	
L46/CML159	-2.5	11.8	**	155.4	10.0		7.8		-5.7		-23.3	-5.9		6.5	-1.3	-0.3		6.8	1.3		-14.8	
L47/CML159	-2.2	8.9	**	139.6	5.1		14.9		8.0		34.5	-0.3		5.7	6.2	-2.0		2.9	-1.7		-10.3	
L48/CML159	-0.1	10.1	**	39.1	-2.4		1.2		-1.6		92.7	-1.0		16.2	0.1	0.2		0.3	4.3		-20.9	*
L49/CML159	-6.5	8.5	**	-87.9	8.4		22.7		12.9		-86.8	-1.0		12.1	0.4	-2.1		-1.2	5.0		-8.7	
L50/CML159	1.4	6.7	**	-120.3	1.6		15.0		11.7		-39.7	-0.3		16.5	-4.8	5.9		3.5	-4.5		-6.1	
L51/CML159	-0.2	6.6	**	-37.3	4.9		20.1		13.8		-73.3	-15.1		0.9	1.5	2.0		4.3	-4.6		-6.7	
L52/CML159	21.4	5.9	**	-137.1	7.6		12.2		1.4		-89.1	-6.1		1.6	1.1	9.1		6.5	0.7		-4.4	
L53/CML159	9.5	6.0	**	88.6	9.5		23.7		14.8		22.8	-0.5		8.6	0.0	2.7		-8.1	-3.6		-3.7	
L54/CML159	2.6	5.6	**	-122.1	4.5		19.6		12.9		-31.8	4.6		5.7	0.4	1.5		-1.9	-3.7		-2.0	
L55/CML159	-2.8	8.7	**	-74.1	-2.7		18.8		21.0	*	-103.4	-5.2		17.8	0.5	-1.1		-0.9	-6.0		-6.7	
L56/CML159	-9.2	5.9	**	-139.5	3.5		20.8		13.8		-70.6	-15.7		21.6	0.0	-1.8		0.1	-5.1		-8.8	
L57/CML159	9.7	6.9	**	-75.0	2.1		22.3		19.2		-70.8	-5.2		-3.0	-4.9	3.4		2.1	-5.9		0.4	
L58/CML159	5.2	6.6	**	-116.1	3.2		18.2		12.2		-89.9	-0.6		-7.8	1.3	-1.5		-0.6	-8.7	*	-2.8	

Appendix 8. Information on single nucleotide polymorphisms (SNPs) used to genotype 58 QPM inbred lines.

No.	Marker	Chrom	Position	base change	Flanking sequence	Locus*
1	PZA00175_2	1	8510027	A/T	TACGGCATCGGCAAC[A/T]TCCTCAGCAGCTTC	AY105791
2	PZA02094_9	1	15725672	A/T	ACAAAGTCTCRCAAT[A/T]AGTACAAAATATAT	AY109844
3	PHM4597_14	1	38608156	A/G	GCTAATATCTGGCGC[A/G]TGCGAGCAGACCGG	N/A
4	PHM11000_21	1	43712019	A/G	TTCGAAGGAGTCAAG[A/G]TATAAATAACAGGG	N/A
5	PZB01062_3	1	56846728	A/G	AAATCCAGAGTTGTG[A/G]TCTTATATACTCCA	FGENESHMAGI_33285.1
6	PZA03561_1	1	60212051	A/C	GGTGCCCTTGACACC[A/C]TCNCTGCTACCAAG	AZM5_85060
7	PZA01267_3	1	76054042	A/G	ACCACTCTGGAGAAA[A/G]MRAAWNNTCAGTTC	AY110690
8	PHM10621_29	1	101421468	C/G	TCCTTCTCAGAGCTA[C/G]CTCTGTCGCTCTCC	N/A
9	PZA01254_2	1	106204446	A/G	GACCTGACTTGATTG[A/G]TCGGCAGTCTGCAG	AY109147
10	PHM1932_51	1	118875639	A/G	TGCCTTGTGGAAGTG[A/G]TCTAACAAGTGCTT	N/A
11	csu1138_3	1	119018614	A/G	CGGAGGGTGGTAGAT[A/G]GGAGTCGGTGATCC	csu1138
12	PHM1725_34	1	150544945	A/C	GACATCTTCGAACAG[A/C]AGCCCAAGTAATGC	N/A
13	PZA02741_1	1	161072169	C/T	CTAATCATCTGAGGC[C/T]AGYGAGGAAGAAGG	AY110942
14	PZA02135_2	1	166550294	A/G	AAAAAAAAGAGTAGC[A/G]CAGGCGGGTTGCTG	AY110206
15	PHM1968_22	1	183647544	A/G	ATGATCGCTTCTTCA[A/G]CTGCTGCCTCCTCC	AY110591
16	PHM5622_21	1	183831657	A/T	TCTGGAGGAAGAAAC[A/T]GAAGGCGATGGCAA	N/A
17	PHM12706_14	1	212356401	A/G	CTCCTCGACCAAGAA[A/G]GCACGAATGATTCT	AY105537
18	PHM1438_34	1	212389447	A/G	CCAGTCAACAGGCAC[A/G]CCATGGATTTGGTT	N/A
19	PZA00664_3	1	227542649	A/G	CACCCTTATGTGATA[A/G]ACGTCGCACAAGTA	AY112024
20	PZA02186_1	1	227896382	A/C	AAATGATACATTAGT[A/C]GTCTCCCCAGTGAT	AY110159
21	PZA02269_3	1	252722026	C/T	GAATTCTTGCTGGAT[C/T]CTGTGTGAATCCTC	AY110621
22	glb1_2	1	257415499	A/T	TGTTGTTKYSTCGTG[A/T]CTAGGCGAGGGAGT	glb1
23	PZB00008_1	1	268371949	A/G	TCCGAACTGGTCGGC[A/G]CCAAATATCGGAGG	BG320409
24	PZB01403_1	1	285273845	A/C	TTCCACCTGTGATGA[A/C]ATTTCCAATCATGT	MAGI_53116
25	PHM4752_14	1	298874066	A/G	GGTGCTGACTACCCA[A/G]GCAATCGTTGTGGA	N/A
26	PZE0186065237	1	NA	T/C	TGATACCATCCCCAA[T/C]CACGCACTGCGTTC	N/A
27	PZE0186365075	1	NA	A/C	AATCGCTARTGCATT[A/C]TCAGGAATCATTCT	N/A
28	PZE-101093951	1	NA	A/G	GAGAGATCCTCACAT[A/G]GCAGCAGATGGCTT	N/A
29	PHM13440_13	2	2527344	A/G	CCAGATGATATACTC[A/G]ATAAGATCGCTCAA	AY112645
30	PZA02337_4	2	15507941	A/G	TGAGTGCTTAGNGCC[A/G]CTGCTGAACCACCT	AY110497
31	PHM6111_5	2	21990814	A/G	ACACAAAGTTGCAGT[A/G]TGCCACACAACCAA	AY107034
32	PZA01374_1	2	28316042	A/G	ATGAGGTTATACGTG[A/G]GCWATCTGCTTAAT	AY104214
33	PZA02378_7	2	35040818	A/G	TGACTGCGACAGGGA[A/G]GGAGGTGCACTTCG	AY110357
34	PZA02496_1	2	40814083	A/C	GAATTACTGAAGACA[A/C]GTAATAAGTTCAGA	AW519877
35	PZA02168_1	2	51780601	C/T	CAGTAGGYAAGACGT[C/T]TGCTCACCTGCATA	AY110275
36	PHM3457_6	2	62804122	A/T	GGATGTCCCAAAGCT[A/T]GACACGATCCTCAC	AY104835
37	PHM13360_13	2	107146579	A/G	CCAGGCATCCATCCA[A/G]CCAACGCTTTGCTA	AY108600
38	PHM3626_3	2	125642617	A/G	GCCATTGACACCAGA[A/G]ATGTGATGCAGATG	AY108532
39	PZA03211_6	2	148837605	C/G	GATGGATATGATCGT[C/G]GAGCGCACGCAGGG	MAGI_108923
40	PZA00495_5	2	170377814	G/T	TGGATGCACTTCGAC[G/T]TTCATCTCCGACCT	AY107322
41	vdac1a_1	2	173767689	C/G	ATAATAAATGAACAT[C/G]TTGGAGGTCCATTT	vdac1a

Appendix 8. Information on single nucleotide polymorphisms (SNPs) used continued

No.	Marker	Chrom	Position	base change	Flanking sequence	Locus
42	PZA03529_1	2	186512251	A/G	GAATAATAACGACAA[A/G]MTGTTAAAAAGRGT	TC310162
43	PHM3668_12	2	195555350	A/T	AAAGCATGAATCATA[A/T]AGTTATGTTGTTTT	AY112434
44	PZB01103_2	2	200020987	A/G	CGTGATGCTGTCACC[A/G]AACTTCAGCAGGCA	1FGENESHMAGI_99686.1
45	PZA01885_2	2	206881202	A/G	TGGCCTCTTATCATC[A/G]CGAGCCTCAACACG	AI668346
46	PZA03602_1	2	209504204	A/T	TCTGGAATTCTGGAT[A/T]TTTATGGGATCGTA	HAM101
47	PZA00527_10	2	216833071	C/T	ACACTCCACTGTAGT[C/T]TAAGGACTGTCAAG	AY112623
48	PZA01991_3	2	220397345	A/G	CCCAGCAATTCTTAT[A/G]CGCCTTGATATCAT	AY109979
49	PZD00022_5	2	233128511	A/C	TATAACYAGGAATCT[A/C]TTCTTCTGTTTGCT	L46400
50	PZA02090_1	3	4138512	A/T	TCTATGGCAGTAAAA[A/T]TCTGCAAGCTGTTT	AY110062
51	PZA00508_2	3	11895613	A/G	NNNNNNNNNNNNNT[A/G]CTAGCATTCAACAG	AY107248
52	PHM2343_25	3	27981649	A/G	AAATAGGTAATGCAG[A/G]CACACCTCTGGGGA	AI664819
53	PZA02255_2	3	33226251	A/C	CCATATTGTAGCACC[G/T]GACAAGCCACAGAG	AY110188
54	PZA01447_1	3	53549251	A/G	AAAATAAATCTGCTG[A/G]GCTGAGGTTGATCA	AY106531
55	PZA02589_1	3	57360226	A/G	CACATCAGTTTCCCC[A/G]TCAGCCAAAGCAGG	AY111731
56	PHM5502_31	3	67284067	A/G	ATGTACACAATCCGG[A/G]AGACTTTCACCCTA	AY106948
57	PZA00109_4	3	82173052	A/T	ACAGTTGCACTTTGA[A/T]TCGACCTTTGGAGA	N/A
58	PZA02742_1	3	97441783	C/G	CCTCACAAGATCCTT[C/G]GCATTTTCCGACAC	AY110942
59	PZA00707_9	3	110715954	A/T	TGTATAGAAGCCAAG[A/T]GCTATGGAAATGGA	AY111703
60	PZA00413_20	3	125192432	A/C	AAGCAAACCATCCTC[A/C]GCACCGGATTCTGC	AY107869
61	PZA02299_16	3	130082791	A/G	GAATAAGCTGCTGCA[A/G]ACTAGAGCCATTAT	AY109541
62	PHM1745_16	3	140079003	A/G	GGAGGGCGTGACATC[A/G]CTGACATGGCTAGG	AY110550
63	PHM4955_12	3	150256198	A/G	TTAACCAAGTGTCAA[A/G]CTACTGCATCTGCC	BE012134
64	PZA00667_2	3	161516227	A/G	GAACGTTGATCTCTG[A/G]GCACACTAATAAAT	AY106743
65	PHM17210_5	3	178229653	A/C	CATCAAACCATCAGA[A/C]TAACAGGCTTTCTG	AY110055
66	PZA03647_1	3	185318331	A/C	GTTTCTCTTCTCGCA[A/C]GCGTTTCGTCTCTC	SDG113
67	PZB01109_1	3	194643731	A/G	TGTGGATAATTGGTG[A/G]GTGGTTTATTTGCT	AZM4_90485
68	PHM15964_16	3	221986592	A/G	CCTTGAATGTGGGTC[A/G]CGTCTTATACACAG	N/A
69	PZA02358_1	4	11329241	A/G	TTAGTTTGTTAAAAA[A/G]NTGTGTTAGCATTC	AY110398
70	PZA02385_6	4	15274450	A/G	TATAYCAGTCCATGC[A/G]TATGACTGCAGAAT	AY110253
71	PZA02457_1	4	29031200	A/G	GGAAGTGGAYAAGGA[A/G]CAGCAGAAGATAGC	AI770588
72	PHM5572_19	4	35384118	A/G	CAATGAAATGGAACA[A/G]GAAACATCGTTTTC	AY107508
73	PZA03247_1	4	35769506	C/G	TCACTATCAGTAATT[C/G]AGTAGCATGTAAGT	M76684
74	PZA00726_10	4	60768063	A/C	CAAGGTTCCAATACC[A/C]CGACGTCAAGAGCA	AY111503
75	bt2_7	4	66290994	A/G	AACCAAGAAGCCAAT[A/G]CCAGATTTCAGGTA	bt2
76	PZA00218_1	4	78946415	A/G	AATATTGGCGGTAAC[A/G]CAAGCTRATGTCGT	AY105676
77	PZA03203_2	4	90203822	A/G	AGGAAGGGGAGATGA[A/G]ATGGCAGATTAAAA	MAGI_106373
78	PZA03536_1	4	107751353	A/G	TATGGAGATACCTMC[A/G]TGCACAAAGGGTTG	TC313290
79	PZA03409_1	4	128632208	A/C	GCTCATCAATTTCTG[A/C]TTTGGTCTGAACAC	AB_486106H08.X2
80	PZA02027_1	4	133043273	C/T	AACTGTGAAATCCTA[C/T]ACCACCCACATCCA	AY109772
81	PZA01477_3	4	172301064	C/T	GTCAACTAGAGTAMT[C/T]GTCGTGGTTCAATG	AY106603
82	PZA00941_2	4	185562016	C/G	AAAGCGTACTGATGY[C/G]TGTCTAAGTATTGT	AY104766
83	PHM4348_16	4	197538207	C/T	TGTATTCACTATTTA[C/T]TACATTGTGATACC	AY111152
84	PZA02779_1	4	207114208	A/G	TTCTCTACAGACTGG[A/G]TGATCTCTGCCGTG	AY110748

Appendix 8. Information on single nucleotide polymorphisms (SNPs) used continued

No.	Marker	Chrom	Position	base change	Flanking sequence	Locus
85	PHM4117_14	4	215393158	A/C	TAGAAGATAAGCTAT[A/C]AGAAATCAAGAACG	N/A
86	PZA00399_11	4	229644826	A/G	CGCGGCTTGATTTCA[A/G]CTGCAACGATGCCG	N/A
87	PZA03322_5	4	242019440	C/G	CACAAATCTGAATTT[C/G]ATCAAACCTCTTTA	ath-miR160
88	PZA00282_19	4	245123514	C/G	TGTTCTATCATCAGA[C/G]RCGAGGCCCTTGTC	AY105452
89	PZA02462_1	5	6820571	A/C	TGACAATTGTGTCCC[A/C]CGCATCTTACACAG	AI881783
90	PZA03092_7	5	11992705	C/T	AAGAATACAAAAACA[C/T]AGCAATGCTGCCTA	AB_486051C10.X2
91	PZA01427_1	5	23135578	A/C	ACTTTTTCAATATKG[A/C]AGATTGAAGAGCAG	AY106398
92	PZA00981_3	5	37030384	A/G	CAATGGAAATGCAGA[A/G]GCAGTCATCCAACA	AY106798
93	PHM12992_5	5	38502372	A/G	AGAGCCATTTTGCTC[A/G]TCGTCTTTCGTCTG	AY111255
94	PZA02207_1	5	49203492	A/G	TCGAAAGATGTTTCA[A/G]CCTCCCATCTGCTA	AY109688
95	PHM4165_14	5	65741535	A/T	CTGTAGATTCATAAT[A/T]GCAGGTGACAAGGG	AY103840
96	PHM16788_6	5	72504523	A/C	TCTTCTTGGTAATAT[A/C]TTGATCCCTTCTCT	N/A
97	PZA00643_13	5	91096945	A/C	TAACTTAGGCAGGAT[G/T]TGCATGGCATGCCA	AY112087
98	PZA00881_1	5	97952638	A/C	TTGCATCAAGAATAC[A/C]TACACATCGATGCC	AY104426
99	PZA02164_16	5	112179855	A/G	CTTGGCCAAGAAGGT[A/G]GCAATGTCGTGRAG	AY109498
100	PZA00636_7	5	115246699	A/G	GTATACATAAGAGTT[A/G]ATCGAACTGACAAG	AY106822
101	PHM662_27	5	135569668	A/C	CCAGTTCCTCGACAC[A/C]GCCTACATCAGCGG	#N/A
102	PZA01983_1	5	135578438	G/T	CAACTCTTATTCCTC[G/T]ATGCAGGGCAAAGA	AY110625
103	PZA01608_1	5	158599491	C/T	GCATAGATTTCTCAC[C/T]GTTTGTCTGTTATT	AY105205
104	ae1_7	5	167873309	A/G	TAATAGGCCATATTC[A/G]TTCTCGGTTTATAC	ae1
105	PHM1899_157	5	179060561	A/G	GCCCTGACTGGCGGT[A/G]TATGTGATCTGCAT	AY107714
106	PZA00352_23	5	191075557	A/G	ATGACACTTTGTTTG[A/G]CAGCTACATGAAGA	AY108254
107	PHM3512_186	5	203434263	A/G	TTGGGAGGGATCACA[A/G]CGCATGCATAATTA	AY105745
108	PZA02480_1	5	214953055	A/G	TCTTCCTCTTGATTC[A/G]YAGCAACCTCAACC	AI901400
109	PZA00440_1	6	22404308	C/G	GCGCGGGTCGTTGGA[C/G]GGACGATGATAGAG	AY107667
110	PZA03120_1	6	57774238	A/C	GACATCAAGCGCGGC[A/C]GGTTCACCGACGAC	MAGI_16437
111	PZA00355_2	6	78756133	C/G	CGAACCAAATGTGAT[C/G]ACAGCAGCTAAYCA	AY108249
112	PZB01009_2	6	84665035	A/C	AGAGAATGAATAAAG[A/C]GGCAGTCTTTGTGC	MAGI_93538
113	PZA00214_1	6	91704092	A/T	GTCAGCACTGAGTCG[A/T]TTGAGGTGCTCGTG	AY109567
114	PZB01658_1	6	102953833	A/T	CCGAGGATAATCGAA[A/T]CAATTCGCGTGGAT	ZMMBLd0003A20.F
115	PZA01029_1	6	114031392	C/T	AATTTGACGTTCTTA[C/T]TGTTTATCGGTTCT	AY107659
116	lac1_3	6	120230802	A/G	GGTCAATCCAGCATT[A/G]TATGTAACACCAAC	lac1
117	PZA00571_1	6	120679943	A/G	GACTGTTGATCTTAT[A/G]GGGGGAGGCTCCCG	AY112197
118	PZA01591_1	6	125113941	A/G	GATCATTTTGTAATG[A/G]TAGTGGCCAATGGA	AY105746
119	PZA01618_2	6	129927781	A/G	AGGGACAACGTGTTC[A/G]ACATGCTCGTCCAG	AY108101
120	PZA02187_1	6	139106115	A/C	GCAGGTAACCTGTAR[A/C]AGTTCCCTCTCACG	AY110137
121	PZA02478_7	6	141111650	A/G	AGTTTGCGCATTCAA[A/G]TCTCGACATGCCCT	AW120211
122	PZA02436_1	6	149251173	C/G	TCCTCCCTCGTCTCT[C/G]ARCAGCACTCATAC	AI665360
123	PZA01462_1	6	155546716	C/T	AAATCACGAAATTTG[C/T]CATCACTATATTTG	AY106576
124	PHM597_18	6	157943848	A/G	GTTCTTGTAGTTCGC[A/G]GAGTCGTCCATGGC	N/A
125	PHM3466_69	6	167148384	A/G	ATCTTCTGAAACAGC[A/G]GCTCAACCCAGGGT	N/A
126	PHM4468_13	6	167527305	A/G	TGATGATGAGAAGAA[A/G]TCTGAGACTGCTAT	AY106902
127	PHM3078_12	7	5963009	A/G	GATACCAGGCGTGAA[A/G]GGGTGCTATGTGGC	AY105159

Appendix 8. Information on single nucleotide polymorphisms (SNPs) used continued

No.	Marker	Chrom	Position	base change	Flanking sequence	Locus
128	PZA02872_1	7	13058813	A/G	CCATCGTGGGTGCCA[A/G]GAAGGACGAGCTCC	AY104876
129	PHM4080_15	7	20240404	A/G	GATGGTGCATGGAAC[A/G]CCTAGGACATAGGA	AY104411
130	PZA03344_2	7	22002659	A/G	GTCCACGCCGGTGGC[A/G]GCGGCGGTAGAGAA	ath-miR172
131	PHM4353_31	7	36392983	A/C	AACCTTCGGCGTAGT[A/C]TGATGTACAGTGAA	AY105616
132	PZA00084_2	7	43948264	C/T	GAACAAATTCGTGCA[C/T]CAGTGAATATAAGG	AY106170
133	PZA03363_1	7	49538583	A/G	GATTTACTTGAATGT[A/G]TAAAAAATACAGTG	AB_605085G01.X2
134	PHM4818_15	7	68051404	C/G	GCTCCATTGCTCCTT[C/G]AGGATATCAAGGCA	AY105836
135	PZA03645_1	7	73892322	C/T	ATAATGTTTCAGTGC[C/T]TATCTGATTGACGA	SDG110
136	PZA01210_1	7	75099046	A/G	GATCAAAACAGCAGT[A/G]CAGTTATCCTTGCA	AY109016
137	PZA01933_3	7	98070498	A/G	CAAAGTCACCAGTGC[A/G]GTGCTAACCATCAT	AW927823
138	PZA01946_7	7	123601837	A/C	ACCATCCAGGCGCGG[A/C]TGCCGCTGCTAATT	BE761686
139	PZD00054_1	7	130488434	A/G	CCATGCCGGCATGAA[A/G]TTATTCATATGCTC	zmm7
140	PZB00752_1	7	131103240	A/C	TGGACCCTGTGCATG[A/C]GCCRGYGGTGGTCC	MAGI_81750
141	PZA00405_6	7	138551416	C/G	CGACCAAATTGAATT[C/G]GTCATTTACGCCAG	AY107911
142	PZA02260_2	7	153012827	C/G	AAGAGGCCTTTTGCT[C/G]TCAAGGTGGATGAA	AY110023
143	PZA00795_1	7	159417489	A/G	AAAATCCTTGGGCGG[A/G]ATACTGAAGATCTT	AY108844
144	PZA01533_2	7	162381818	A/G	TGTTGCATTTGAGCT[A/G]YTRCGCTAGTTAAT	AY105005
145	PZA02174_2	8	4101256	G/T	TGTTAAAGGAGCTTA[G/T]ATCAATTTAAGGAG	AY110581
146	PZA02955_3	8	14721554	A/C	ACTGTCTCCGTGATC[A/C]ACTGCGGCACCTAA	AY104508
147	PHM2350_17	8	23985819	A/C	CAATATGTTTGTGTT[A/C]TGCAGACCTTCAAT	AY108896
148	PZA00498_5	8	48775713	A/C	GAAACAGTGGTACAA[A/C]TACGCCACCAACAG	AY107308
149	PHM3856_10	8	55008368	A/G	TTGCCCCTTGCGATT[A/G]ATTGTGTGCCGCTT	N/A
150	PZA00793_2	8	64421988	A/C	AGTGTACAAAAAGCC[A/C]ATTGGGAATGTTCA	AY108840
151	PZA00379_2	8	65657055	C/T	CAGAGCCTACCTGGA[C/T]GGTGCCCAGGTTGG	AY108089
152	PHM11114_7	8	70899841	A/G	CAACCGAAACCGGCA[A/G]GTCTGACATGTCTA	AY105907
153	PHM4134_8	8	105795742	C/G	GACGGAGAGGCTAGT[C/G]GTTATGAGGGGCGA	AY108545
154	PZA01972_14	8	111333947	C/G	AACCAAGTTCAGCGA[C/G]GTTCTTGTTCTGAG	AY110056
155	PHM5805_19	8	120875248	A/C	GCCAATTTCTTTGGT[A/C]GTATATCTGGGCTG	N/A
156	PZB02155_1	8	123272935	A/C	CATGATGCATGGCTG[A/C]CATATCTGGCGTCC	MAGI4_124049
157	PZA00770_1	8	134140609	C/G	TCTAGTACATCTGGG[C/G]GGTTTGTCATACAT	AY104797
158	PZA02011_1	8	140212456	A/G	TTAGYGKGTGCTGTC[A/G]TTTTCTATGCATCA	AY109767
159	PZA03651_1	8	146342784	A/C	TACCTGGAATTAACA[A/C]AGCAAATAACATGA	SDG115
160	PZA03182_5	8	152155087	C/T	TGGGGAACAGGTACG[C/T]ACAATGCGTTCCGG	MAGI_60276
161	PZA01857_1	8	156100505	A/G	CCCAAAGCTGCTGAA[A/G]TTTGTTGATCCAAC	N/A
162	PZA02746_2	8	163067200	G/T	TGTTCACAGCCTTCC[G/T]TGGGTTTTTGAGAG	AY112084
163	PHM2749_10	8	171703522	A/G	CAAGAGGATGAGAGC[A/G]CACTATGAAGTCCT	AY107094
164	PHM4942_12	9	3166446	A/G	GAAACAACAAATTTA[A/G]GCATCGAAGTATAT	AY111813
165	sh1_12	9	11340882	A/G	CATGTCTGCTCCAGG[A/G]GAGACAATGTTGAA	sh1
166	PHM5181_10	9	15582065	C/T	CTCAACCAGCACAGG[C/T]ACAGGCTAGTTCAT	AY104700
167	PZA00860_1	9	18684111	A/C	TTATGTAATGGGTWT[A/C]AGCAGGATTAATCA	AY104369
168	PZB01110_6	9	24028336	A/G	CTCCTCCCGTACTCT[A/G]GAATCTTCTCGCCC	AZM4_91097
169	PHM229_15	9	30003189	A/G	CTGGCAGGAGCAATC[A/G]CCCTAGGTGAATCA	N/A
170	PZA01791_2	9	77467426	C/T	TGTTTGCCACTTAAG[C/T]AGCTAAATGGATGA	AY106098

Appendix 8. Information on single nucleotide polymorphisms (SNPs) used continued

No.	Marker	Chrom	Position	base change	Flanking sequence	Locus
171	PZB00761_1	9	83959651	A/G	AGTGCGGCTCAAGTA[C/T]GTGAAGCTGGGGTA	MAGI_38349
172	PZA01062_1	9	88057320	A/C	GGTCTTGGATAATAA[A/C]AACACCAGCAAAGG	AY107743
173	PZA03596_1	9	90436248	C/T	ACAGATACAACTCAA[C/T]GCACACATATACAA	HAF101
174	PZB01899_1	9	98502843	A/G	GAACTTGTACTGCGC[A/G]CTCCACCGCTACTC	MAGI4_79260
175	PZB01358_1	9	106774837	A/G	AAATGCATATCAACG[A/G]AAGAACAATCTCTT	MAGI_32337
176	PZA02325_4	9	117870773	A/T	CATGTACATATANTA[A/T]TAATAGTTGGAACA	AY109764
177	PZA00840_1	9	124112589	A/C	TTCGTTGTGTRRGGG[A/C]TGAGTGGTTTGTCG	AY104332
178	PHM7916_4	9	132762904	A/G	CTCTTCACTGTCAGC[A/G]TCACCATCGACCTC	N/A
179	PZA01096_1	9	133450713	G/T	ATTCTTTGGACAAGA[G/T]GTATACTATTACAG	AY107103
180	PZA01715_2	9	142948449	C/T	ACAAATGATATATAT[C/T]GGTATATGTATTCC	AY108278
181	PZA01715_1	9	142948545	A/G	CCAATCCAAGCTGGG[A/G]TAGGCAAAGTAGAG	AY108278
182	PHM1911_173	9	148570003	A/G	AGCTCTGGTTGCACG[A/G]TCGAGTTAACCGGT	AY103907
183	PZA01313_2	10	3598262	A/G	AAGTACGGTTGCAAA[A/G]CAAGAATTTGACTA	AY110922
184	PHM2828_83	10	6121374	C/T	ATTATGTTATCGTCA[C/T]TGGCTGTACCTTTG	AY103908
185	PHM1752_36	10	9746552	A/G	ACGTGTGTACTGATC[A/G]TGGTTAAGCACGTA	N/A
186	PHM15331_16	10	10432605	A/G	CGTCTTCAGGCGCTC[A/G]CTGCGGTTGGACGT	AY108721
187	PHM3922_32	10	17722938	A/G	GACACACACGAACAT[A/G]TACACACATACAAG	AY107581
188	PHM4066_11	10	41187565	A/G	TGGACCACATCCATC[A/G]CTTTCTTCTTCGCA	N/A
189	PHM1155_14	10	62063210	A/G	GCCAACTGGCCAGTT[A/G]GAGAAGGTGCCCTT	N/A
190	PHM2770_19	10	72565410	A/C	AGCTGCTTTAGAAAT[A/C]TCTCCCTCCAATTC	AY110321
191	PZA00814_1	10	87194491	A/C	CCATCACCAAGAACA[A/C]CTAGCTAGTCATCC	AY111401
192	PZA01919_2	10	111260278	C/G	GTGATCAACATAAAA[C/G]CATCCATTCTTGTA	AW244184
193	PHM13687_14	10	117796822	A/G	AGTATAACAATGGAT[A/G]TAGATCTTCTGAAC	AY111609
194	PZA03713_1	10	121489957	A/T	TTGGTCCTGTCTTTA[A/T]TGATAGCTGCGAAA	AJ400868
195	PZA00866_2	10	124203565	A/G	TGGTAGAGGAATCTG[A/G]TGACTGGCTGCGGT	AY111873
196	PZA01241_2	10	130452676	A/C	TTAAGTACAAAAGTA[A/C]AGCCTTAGCTGTCA	AY109120
197	PHM15868_56	10	137132183	A/G	ATGGAGCAGTAAGCT[A/G]GAGTGCGAGTTGGA	AY107531
198	PZA03605_1	10	141830532	A/G	ATAAGTTTGACACTT[A/G]TAAAATGTGCATGT	DMT102
199	PHM3309_8	10	144752780	A/G	CCAAGGGCCTGAAGG[A/G]TACAGAGGGATTTA	N/A
200	PHM3736_11	10	147762925	A/G	GAAGAGCAGTGAGAA[A/G]CTGGTGAGGAGATT	N/A
201	opaque2-SNP1	7	NA	T/C	GCACACCGCCGCCGG[T/C]GGTGGTGGTGCCGA	N/A

**Locus information found from McMullen et al. (2009)*